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STUDIES ON PHALAENOPSIS, III

P. EQUESTRIS (SCHAUER) REICHB. F., P. LINDENII LOHER P. LUEDDEMANNIANA REICHB. F., P. MARIAE BURB. AND P. MICHOLITZII ROLFE

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FIVE PLATES

This paper is the third series on studies on Philippine species of *Phalaenopsis*,[1] under the sections *Zebrinæ* and *Stauroglottis*. It comprises the following species: *P. equestris* (Schauer) Reichb. f., *P. Lindenii* Loher, *P. Lueddemanniana* Reichb. f., *P. Mariae* Burb., and *P. Micholitzii* Rolfe. Many years of study of Philippine orchids gave me an opportunity to restudy the above species in their living conditions, particularly the Reichenbach's species. This paper includes also a brief discussion of excluded and doubtful species. The following are excluded for two reasons: (*a*) species which were erroneously credited to the Philippines, and (*b*) species which have not been seen by the author.

Various sections of *Phalaenopsis* have been proposed. Pfitzer [2] proposed five sections, of which three are represented in the Philippines (*Euphalaenopsis, Zebrinæ,* and *Stauroglottis*). The two other sections (*Proboscidioides* and *Antenniferæ*) are also represented but by introduced species.

Rolfe [3] has proposed the sixth section (*Esmeralda*), which is represented in the Philippines by introduced species, and which is no different from Pfitzer's *Antenniferæ.*

[1] Previous papers. I: Phil. Jour. Sci. 74 (1941) 175–185, *2 plates;* II: Phil. Jour. Sci. 76 (1941) 81–97, *9 plates.*

[2] Pfitzer, in Engl. & Prantl, Pflanzenfam. II 6 (1889) 212.

[3] In Veitch, Man. Orch. Pl. pt. 7 (1891) 17.

Key to the sections of Phalaenopsis.

1. Petals much broader than sepals and contracted at the base.
   2. Middle lobe of lip with two cirrhi or two divaricate lobes at the apex; without proboscislike rostellum..... *Euphalaenopsis.*[4]
   2. Middle lobe of lip without apical appendages; with proboscislike rostellum ..... *Proboscidioides.*[5]
1. Petals equal to, rarely smaller than, sepals; middle lobe of lip entire, without apical appendages and without proboscislike rostellum.
   2. Claw of lip without appendages.
      3. Middle of lobe of lip ovate; upper surface smooth.... *Stauroglottis.*[6]
      3. Middle lobe of lip oblong; upper surface with a crest of hairs. *Zebrinae.*[7]
   2. Claw of the lip with a pair of slender appendages..... *Antenniferæ.*[8]

Section STAUROGLOTTIS Schauer

Sepalen und Petalen siemlich gleich, meist 1 farbig, Endlappen der Lippe ungeteilt, quer verbreitert, oft am Grunde mit zahlreichen fadigen Forstsatzen, z. B. *Ph. Parishii* Rchb. f. aus Birma.[9]

Key to the Philippine species.

1. Leaves green; middle lobe of lip ovate..... 8. *P. equestris.*
2. Leaves marbled and barred with silvery gray; middle lobe of lip suborbicular ..... 9. *P. Lindenii.*

PHALAENOPSIS EQUESTRIS (Schauer) Reichb f. Plate 1, fig. 1; Plate 2.

Phalaenopsis equestris (Schauer) REICHB. f. in Linnæa 22 (1849) 864; LINDL. in Paxt. Flow. Gar. 2 (1852) 174; REICHB. f. in Walp. Ann. 3 (1852) 562; 6 (1864) 860; MIQ., Fl. Ind. Bat. 3 (1859) 690; REICHB. f. in Hamb. Gartenz. 16 (1860) 116; DUCHARTRE in Jour. Soc. Imp. et Centr. Hort. Par. 6 (1860) 869, 8 (1862) 727; REICHB. f., Xen. Orch. 2 (1862) 4; NAVES, Novis App. (1882) 242; AMES, Orch. 2 (1908) 229, 5 (1915) 216, ex Merr. Enum. Phil. Fl. Pl. 1 (1925) 413; SCHLECHTER, Die Orchideen (1927) 537.

[4] Section proposed by Bentham. Philippine species under this section published in Phil. Jour. Sci. 74 (1941) 175–187, two plates; Phil. Jour. Sci. (1941).

[5] Section proposed by Pfitzer, in Engl. & Prantl, Pflanzenfam. II 6 (1889) 212; typified by *P. Lowi* Reichb. f.

[6] Section proposed by Schauer [*see* Engl. & Prantl. Pflanzenfam. II 6 (1889) 212]; typified by *P. Parishii* Reichb. f., and by *P. equestris* (Schauer) Reichb. f.

[7] Section proposed by Pfitzer, loc. cit.; typified by *P. Lueddemanniana* Reichb. f.

[8] Section proposed by Pfitzer, in 1889, which was based on *P. antinnefera* Reichb. f. which is now made a synonym of *P. esmeralda* Reichb. f. (1874). According to Veitch [Man. Orch. Pl. pt. 7 (1891) 17] Rolfe proposed the section *Esmeralda* for species with a pair of slender appendages in the claw of the lip. Section *Esmeralda* was, therefore, proposed 17 years after Pfitzer had proposed the section *Antenniferæ.*

[9] Pfitzer, loc. cit. 212.

Stauroglottis equestris SCHAUER in Nov. Act. Acad. Nat. Cur. 19 Suppl. 1 (1843) 432.

Phalaenopsis rosea LINDL. in Gard. Cron. (1848) 671, *text cut;* Paxt. Mag. Bot. 16 (1849) 60, 189, *text cut;* LINDL. in Paxt. Flow. Gard. 2 (1852) 173, *t. 72;* REICHB. f. in Bot. Zeit. 10 (1852) 673; MOORE, Ill. Orch. Pl. (1857) Phalaen. 7; HOOK. in Bot. Mag. 86 (1860) *t. 2512;* LEM. in Jard. Fleur. 3 (1853) *t. 283,* in Ill. Hort. 10 (1863) Misc. 11; VAN HOUTTE in Fl. des Serres 16 (1866) t. *1645;* JENNINGS, Orch. (1875) *t. 27;* BURB. in The Garden 22 (1822) 119 (excl. var.); VIDAL, Phan. Cuming. Philip. (1885) 150, Rev. Pl. Vasc. Filip. (1886) 270; ROLFE in Gard. Chron. II 26 (1886) 276; WARNER & WILL., Orch. Alb. 6 (1887) *t. 268;* VEITCH, Man. Orch. Pl. pt. 7 (1891) 34; AMES, Orch. 1 (1905) 102.

Phalaenopsis rosea Lindl. var. *leucaspis* ROLFE in Gard. Chron. 26 (1886) 276; VEITCH, Man. Orch. Pl. pt. 7 (1891) 34.

Phalaenopsis esmeralda COGN. in Dict. Icon. Orch. (1898) Phalaen. *t. 3,* non Reichb. f.

Phalaenopsis equestris (Schauer) Reichb. f. var. *leucaspis* REICHB. f. in Gard. Chron. II 15 (1881) 688, in l'Orchidoph. 1 (1881) 50; AMES, Orch. 2 (1908) 230.

Phalaenopsis equestris (Schauer) Reichb. f. var. *leucotanthe* REICHB. f. in l'Orchidoph. 3 (1883) 490; AMES, Orch. 2 (1908) 230; AMES & QUIS. in Phil. Jour. Sci. 52 (1933) 454, *t. 2, figs. 7–8; t. 11, fig. 2.*

The original description reads as follows:

Stems very short. Roots greenish or purplish, fleshy. Leaves fleshy, light green or dull green, 2 to 4, oblong, elliptic-oblong or oblong-obovate, usually 10 to 15 cm, up to 21 cm long, 3 to 5 cm wide, the apex recurved, subacute or obtuse, slightly narrowed to the base. Scapes lateral, arising from between the lower leaves, simple or branched, 15 to 47 cm long, few- or many-flowered; the rachis purplish, terete. Flowers odorless, 2.5 to 4 cm across. Pedicellate ovary slender, white with pale green at the base, 1.5 to 1.9 cm long. Sepals and petals spreading, nearly equal in size and shape, white flushed with rose purple at the center and especially near the base. Sepals oblong-lanceolate, 13 to 14 mm long, 6 to 7 mm wide, the apex obtuse, and rather broad at the base. Petals narrowly rhomboidal, obtuse, 13 to 14 mm long, 8 to 9 mm wide, somewhat constricted at the base. Labellum tri-lobed; middle lobe ovate, acute or briefly acuminate, fleshy, entire, without apical appendages, with a depression at the middle, 11 to 12 mm long, 8 to 9 mm wide, rose purple, darker purple at the tip and flushed with little orange at the base, the margins often reflexed; lateral lobes small, linear-spathulate, oblique, recurved, 6 to 8 mm long, 2 to 2.5 mm wide at the widest portion, white flushed with pale rose purple, often streaked with purple lines within. Callus fleshy, subquadrate, white, or yellow dotted with flame scarlet or morocco red. Column terete, curved slightly, white with rose purple above, 8 to 9 mm long, the beak long and white. Anther cap broadly ovate. Pollinia 2, ellipsoid, cream-colored. Capsules linear, 6 to 7 cm long, excluding the pedicels (1.5 to 2 cm long), 0.5 to 0.8 cm in diameter.

PHILIPPINES, without locality, *Cuming 2051* (in herb. Brit. Mus.; specimen not seen). BATAN ISLAND, Mt. Iraya, *Bur. Sci. 80793 Ramos.* LUZON, Ilocos Norte Province, Bangui, *Bur. Sci. 7736, 27618 Ramos;* without locality, *Lyon 3401:* Isabela Province, Palanan Bay, *Bur. Sci. 21168 Escritor:* Bataan Province, Mt. Mariveles, *Elmer 6861, Williams 376, For. Bur. 2280 Meyer, Merrill 3849;* Lamao, *Bur. Sci. 3043, 5605 Cuzner, Bur. Sci. 1895 Foxworthy:* Rizal Province, without locality, *Loher 3532;* Jalajala, *Bur. Sci. 11931 Robinson & Ramos;* Antipolo, *Bur. Sci. 49637 Ramos:* Manila, *Bur. Sci. 85571 Quisumbing* (living plants from Rizal Province, typical of var. *leucotanthe* Reichb. f.): Laguna Province, Santa Maria–Mabitac, *For. Bur. 8906 Curran:* Tayabas Province, Mt. Tulaog, *Ramos & Edaño, s. n. 1917;* Casiguran, *Phil. Nat. Herb. 3230 Vanoverbergh;* Mt. Pular, *Bur. Sci. 19408 Ramos;* Guinayangan, *Bur. Sci. 20775 Escritor:* Camarines Sur Province, without locality, *For. Bur. 22628 Alvarez, For. Bur. 12283 Curran:* Albay Province, Mayon Volcano, *Bur. Sci. 2381 Mearns.* BOHOL, *Bur. Sci. 1235 McGregor.* MINDANAO, Davao Province, Baganga, *Rev. R. F. Black 26;* Todaya, *Copeland 1228:* Lanao Province, Camp Keithley, *Clemens 5622.* CAMIGUIN ISLAND, Mambajao, *Elmer 14247.* The species have been reported also from the islands of Samar, Leyte, Negros, Cebu, and Panay; no records from Palawan or Mindoro. A common and widely distributed species, altitude from sea level to 300 meters. It is called in English "Rose colored Phalaenopsis," and locally "rosea." The plant flowers throughout the year, but more profusely during February to May. This species is peculiar like other *Phalaenopsis* in producing young plants on the old stems and old roots. Scapes need not be cut after flowering as from these old ones new branches are developed producing flowers. The species is endemic.

Two varieties have been recognized by Reichenbach f. (*leucaspis* and *leucotanthe*); *leucaspis* differing from the species in its smaller flowers and in having more deeply colored midlobe of the lip; and *leucotanthe* differing in the color of flowers being white. The differences being in color only, the two varieties have not been recognized in this paper.

Phalaenopsis equestris is a typical representative of the section *Stauroglottis.* The species is characterized by its light-green or dull-green leaves, some forms resembling those of *P. aphrodite.* The flowers are small, with petals and sepals with

practically the same color and shape, usually white, flushed with rose purple. The labellum is trilobed, with the middle lobe ovate, entire, and without appendages.

PHALAENOPSIS LINDENII Loher. Plate 1, fig. 1; Plate 4, figs. 1–9; Plate 5.

Phalaenopsis Lindenii LOHER in Jour. des Orch. 6 (1895) 103; Orchis 1 (1907) 82, *fig. 37;* ROLFE in Orch. Rev. 13 (1905) 230, 15 (1907) 296; AMES in Phil. Jour. Sci. 4 (1909) Bot. 599, Orch. 5 (1915) 217, ex Merr. Enum. Phil. Fl. Pl. 1 (1925) 414; G. WILSON in Orch. Rev. 30 (1922) 354.

The original description reads as follows:

Phalaenopsis Lindenii Loher.—Cette nouvelle espéce est dédicé á M. J. Linden par l'explorateur que la découverte, et qui en donne la déscription suivante:

Folia oblonga, albido-argentea, viridi-maculata; pedunculi purpurei, bracteis parvis, acutis; perigonii phylla exteriora et interiora subaequalia, obovata subclavata, oblusa, albida (versus nervum medium subrosea); labelli tripartiti lobi laterales subfalcati, oblongi-obtusi, versus basin interionem maculis aurantiacis, scatello vel callo bilobo aurantiaco maculato; lobus intermedius cordato-rotundatus breviter acuminatus, striis quinque purpureis, basi albidus, medio superiori amethystinus.

Cette espéce rappelle un peu par son feuillage le *P. Schilleriana* mais elle a les feuilles beaucoup plus étroites, á peu prés gladiolées; quant aux fleurs, elles se rapprochent beaucoup a celles du *P. rosea*, mas elles sont beaucoup plus grandes, presque doubles. En oulre, elle s'en distinguent par let coloris du labelle, qui a le lobe antérieur améthyste vif avec la base rose pale; cet organe est sensiblement arronde, briévement acuminé tandis que dans le *P. rosea* il a la forme d'un losange.

M. Loher remarque qu'aucun autre Phalaenopsis ne croit dans l'endroit ou se rencontre la nouvelle espéce.

Habit similar to *P. equestris.* Leaves oblanceolate or narrowly oblong-oblanceolate, subacute, 17.5 to 20 cm long, 2.5 to 4 cm wide, deep dull green, marbled and maculated with silvery gray above, purplish beneath (resembling somewhat thin leaves of *P. Schilleriana*). Scapes few-flowered, simple or branched, much longer than the leaves, 20 to 50 cm long. Flowers odorless, 3 to 3.5 cm across. Pedicellate ovary, slender, 2 to 3 cm long. Sepals and petals white, flushed with light rose purple, each marked with 5 to 7 defined purple lines. Dorsal sepal oblong-elliptic, obtuse, 14 to 15 cm long, 6 to 8 mm wide. Lateral sepals oblong-ovate, falcate, obtuse, 14 to 17 mm long, 7 to 9.5 mm wide. Petals obovate-spathulate, broadly obtuse, 13 to 15 mm long, 8 to 10 mm wide at the widest portion. Labellum trilobed; middle lobe suborbicular, apiculate, 10 to 12 mm long, 9 to 12 mm wide, mallow purple with 5 or 7 well-

defined radiating rhodamine purple lines, the base and apiculum white; lateral lobes narrowly oblong, subspathulate, dilated at the apex, obtuse, 7.5 to 9 mm long, 2.5 to 3 mm wide, white, flushed with phlox purple at the apex, and dotted with ferruginous at the base. Column terete, 7 to 9 mm long, white, the anterior surface rhodamine purple. Callus disc-shaped when spread out, white dotted with ferruginous. Anther cap broadly ovate. Pollinia two, ellipsoid.

LUZON, Benguet subprovince, Baguio, *For. Bur. 5121, 5122 Curran, Williams 1947 bis, Phil. Nat. Herb. 7984 Quisumbing.* The species is endemic. It occurs at higher altitudes. It flowers from March to August.

Phalaenopsis Lindenii is perhaps a natural hybrid between *P. equestris* and *P. Schilleriana.*

Rolfe [10] suspected it also to be a natural hybrid of the two species mentioned. The marbled and maculated leaves except size and shape suggest those of *P. Schilleriana,* though the leaves of this species are more delicate and thinner. The flowering habit is that of *P. equestris.* The general habit of growth, size of flowers, details of the flowers except the middle lobe of the lip suggest those of *P. equestris.* The absence of *P. Schilleriana* in regions where this species grows is rather weak argument in favor of the parentage of this species. It is, however, possible that *P. Schilleriana* may have existed in these regions where *P. Lindenii* now grows. We have a parallel case of *P. Schilleriana-Stuartiana* and *P. aphrodite* var. *Sanderiana* of Mindanao. Whether the species in question is a natural hybrid or not, it is conclusive that *P. Lindenii* is a distinct species. It is closely allied to *P. equestris,* differing markedly in its marbled and maculated leaves, and the shape of the middle lobe of the lip. It is not allied to *P. Schilleriana* because of the absence of apical appendages at the middle lobe of the lip. The species was dedicated to Mr. M. J. Linden.

Section ZEBRINAE Pfitzer

Sepalen und Petalen ziemlich gleich, meistens mit farbigen Querbändern auf hellem Grund, Endlappen der Lippe ungeteilt, länger als breit. Hierber *Ph. sumatrana* Korth. Rechb. f. aus Sumatra und *Ph. Luddemanniana* Rchb. f. von den Philippinen, beide oft gezogen, sowie *Ph. speciosa* Rchb. f. (Fig. 213 links).—PFITZER, loc. cit. 212.

Leaves green. Middle lobe of the lip longer than wide, the upper surface with a crest of hairs; petals and sepals barred.

[10] Orch. Rev. 13 (1905) 230.

Typified in the Philippines by *Phalaenopsis Lueddemanniana* Reichb. f.

Key to the Philippine species.

1. Labellum oblong or oblong-oblanceolate.
   2. Flowers 4 to 5 cm across; dorsal sepal oblong or oblong-elliptic, acute ..... 10. *P. Lueddemanniana.*
   2. Flowers smaller, not more than 3 cm across; dorsal sepal narrowly oblong, obtuse..... 11. *P. Mariae.*
1. Labellum rhombic-spatulate..... 12. *P. Micholitzii.*

PHALAENOPSIS LUEDDEMANNIANA Reichb. f. Plate 1, figs. 3–6; Plate 3.

Phalaenopsis Lueddemanniana REICHB. f. in Bot. Zeit. 23 (1865) 146, in Gard. Chron. (1865) 434; MOORE in Flor. & Pomol. (1865) 257, *t. 254;* LEM. in Ill. Hort. 12 (1865) Misc. 31; EDIT. in Proc. Roy. Hort. Soc. 5 (1865) 137; OTTO in Hamb. Gartenz. 21 (1865) 470; G. B. in Belg. Hort. 15 (1865) 229; CARR. in Rev. Hort. 44 (1872) 390, *t.;* ROLFE in Gard. Chron. II 26 (1886) 277, in Lindenia 2 (1886) 95, *t. 94,* 8 (1892) 63, *t. 366;* VEITCH, Man. Orch. Pl. pt. 7 (1891) 30, *text cut;* COGN. in Dict. Icon. Orch. (1899) Phalaen. *t. 9;* AMES, Orch. 2 (1908) 230, 5 (1915) 217, ex Merr. Enum. Phil. Fl. Pl. 1 (1925) 415.

Phalaenopsis Lueddemanniana Reichb. f. var. *delicata* REICHB. f. in Gard. Chron. (1865) 434; BURB. in The Garden 22 (1882) 119; ROLFE in Gard. Chron. II 26 (1886) 277, in Lindenia 8 (1892) 63, sub *t. 366;* AMES, Orch. 2 (1908) 231.

Phalaenopsis Lueddemanii BOXALL ex Naves, Novis. App. (1882) 243, sphalm.

Phalaenopsis Lueddemanniana BATEM. in Bot. Mag. 91 (1865) *t. 5523,* Second Cent. Orch. Pl. (1867) *t, 133,* non Reichb. f.; VAN HOUTTE in Fl. des Serres 16 (1865) 53, *t. 1636.*

Phalaenopsis Lueddemanniana Reichb. f. subvar. *delicata* VEITCH, Man. Orch. Pl. pt. 7 (1891) 30.

Phalaenopsis Lueddemanniana Reichb. f. var. *hieroglyphica* REICHB. f. in Gard. Chron. III 2 (1887) 586; EDIT. in l'Orchidoph. 9 (1889) 197; ROLFE in Lindenia 8 (1892) 63, sub. *t. 366;* AMES, Orch. 2 (1908) 231.

Phalaenopsis Lueddemanniana Reichb. f. subvar. *hieroglyphica* VEITCH, Man. Orch. Pl. pt. 7 (1891) 31.

Phalaenopsis Lueddemanniana Reichb. f. var. *ochracea* REICHB. f. in Gard. Chron. (1865) 438; CARR. in Rev. Hort. 44 (1872) 391, *fig. A;* BURB. in The Garden 22 (1882) 119; ROLFE in Gard. Chron. II 26 (1886) 277, in Lindenia 8 (1892) 63, sub. *t. 366;* AMES, Orch. 2 (1908) 232.

Phalaenopsis Lueddemanniana Reichb. f. subvar. *ochracea* VEITCH, Man. Orch. Pl. pt. 7 (1891) 31.

Phalaenopsis Lueddemanniana Reichb. f. var. *pulchra* REICHB. f. in Gard. Chron. II 4 (1875) 36; BURB. in The Garden 22 (1882) 119; ROLFE in Gard. Chron. II 26 (1886) 277, in Lindenia 8 (1892) 64, sub. *t. 366;* AMES, Orch. 2 (1908) 232.

Phalaenopsis Lueddemanniana Reichb. f. subvar. *pulchra* VEITCH, Man. Orch. Pl. pt. 7 (1891) 31.

Phalaenopsis Lueddemanniana Reichb. f. var. *purpurea* AMES & QUIS. in Phil. Jour. Sci. 49 (1932) 494, *t. 2, 10, 24.*

Phalaenopsis Boxallii REICHB. f. in Gard. Chron. II 19 (1883) 274; ROLFE in Gard. Chron. II 26 (1886) 276; VEITCH, Man. Orch. Pl. pt. 7 (1891) 26; AMES, Orch. 5 (1915) 216, ex. Merr. Enum. Phil. Fl. Pl. 1 (1925) 413.

The original description reads as follows:

Phalaenopsis Lueddemanniana aff. *Ph. sumatranae* Korth. et Rchb. fil. (*zebrinae* Hort. Bog.), et *violaceae* Teism. et Binnd. sepalis tepalisque cuneato-oblongis acutis, labello tripartito, partitionibus lateralibus ligulatis, apice excisobidentatis, extus medio umbonato carinatis erectis, partitione media ab ungue angusto oblonga antice apice utrinque angulata, sen dentata, seu serrata, fornicata ante basin ac apice carinata, carinis nunc serratis, antice pilis circumdata, papulis seriatis as ligulis bifidis duabus a disco inter partitiones posticas in basin partitionis mediae, columna utrinque basi angulata.

Diese Art blünte zuerst bei Herrn Lüddemann in Paris (Boulevart des Gobelins), der sie von den Philippinen einführte. Sie ist eine sehr schöne Pflanze. Die Lippe und Säule sind amethystfarbig. Die Sepalen und Tepalen ebenso und mit vielen braunen Querbinden.

Ein herrliches Exemplar mit grossen zungigen Blättern und einem dreiblüthigen und einem einblüthigen Blüthenstiel sah ich bei Herrn Dr. Pattison in London, S. Johns Wood, 10. Cavendish road. Ferner sah ich die Pflanze in Blüthe bein Herrn Day, High Cross, Tottenham und in Knospen bei Herrn Low, Upper Clapton.

Auf alle Fälle ist sie eine glänzende Acquisition für unsere Gärten. Ich lasse dahin gestellt, ob nicht einmal Mittelformen sich zeigen werden, welche die Vereinigung mit den obengenannten zwei Arten nöthig machen, was indessen nicht sehr wahrscheinlich.—REICHB. F., Bot. Zeit. 23 (1865) 146.

Stems short. Roots greenish. Leaves 3 to 5, somewhat shining, fleshy but not as fleshy as *P. amabilis*, pale green or yellowish green, oblanceolate or oblong-oblanceolate, 10 to 15 cm long, in some forms up to 33 cm long, 3.5 to 5 cm wide, in some cases up to 7.5 cm wide. Scape few-flowered, usually unbranched, 6.5 to 10 cm long, up to 30 cm sometimes; peduncles greenish. Flowers usually odorless, in some forms particularly the Sorsogon form, fragrant, 4 to 5 cm across. Pedicellate ovary slender, pale green, 2 to 3 cm long. Sepals and petals spreading, white or yellowish background, sometimes suffused with phlox purple, and marked with transverse bars of amethyst purple (in some forms with ferruginous bars). Dorsal sepal oblong or oblong-elliptic, acute, 2 to 3 cm long, 1 to 1.5 cm wide. Lateral sepals oblong or oblong-ovate, falcate, acute, 2.2

to 3 cm long, 1 to 1.5 cm wide. Petals slightly smaller than the sepals, elliptic-ovate, acute, somewhat constricted at the base, 2 to 3 cm long, 1 to 1.3 cm wide. Labellum fleshy, trilobed; middle lobe narrowly oblong or oblong-oblanceolate, entire, 1.3 to 1.5 cm long, 0.6 to 0.8 cm wide at the widest portion, white or purplish, with the tip greenish, with a crest of white hairs on the surface (these limited or extended), and a thin keel at the base; on the disk between the lateral lobes are a series of minute fleshy scales (few or many) with two forcepslike appendages in front, these white or phlox pink; lateral lobes erect, ligulate, typically double-toothed at the apex (we have a series from simple without tooth to deeply double-toothed), 6 to 7 mm long, 2.2 to 3 mm at the base, white with mallow pink or orange near the base. Column terete, clavate, white, the base light phlox purple, 12 to 13 mm long. Anther cap ovate, pale lumiere green. Pollinia two, ellipsoid.

LUZON, Nueva Vizcaya Province, Dupax, *Bur. Sci. 11136, 11141 McGregor:* Pangasinan Province, Mt. Isidro, *For. Bur. 8362 Curran & Merritt:* Bulacan Province, Norzagaray, *Bur. Sci. 13046 Ramos:* Manila, cultivated, *Bur. Sci. 84548, 84549 Quisumbing* (living plants from Mt. Mariveles, Bataan Province) : Rizal Province, Pasay, cultivated, *Phil. Nat. Herb. 8079 Quisumbing* (living plants from Montalban, Rizal Province) ; without locality, *Loher 14650, Bur. Sci. 3069 Ramos:* Laguna Province, San Antonio, *Bur. Sci. 20443 Ramos, For. Bur. 19272 Curran, Loher 6005:* Tayabas Province, Mt. Binuang, *Bur. Sci. 28551 Ramos & Edaño;* Mt. Pular, *Bur. Sci. 19364 Ramos:* Sorsogon Province, Mt. Bulusan, *Elmer 15768.* POLILLO (Tayabas Province), *Bur. Sci. 10437 McGregor.* LEYTE, Tacloban, *For. Bur. 12452 Danao.*

A common and widely distributed species, epiphyte, at low altitude to 60 meters.

Phalaenopsis Lueddemanniana is a variable species, particularly in color. While in the typical forms the sepals and petals are transversed by bars of amethyst purple, in some other forms these bars are ferruginous and in others purplish with no bars; the background may be white or yellowish. As the differences between *P. Boxallii* and this species are merely in the color of the flowers, *P. Boxallii* is reduced to synonymy. There are five varieties which have been described; but as the differences are in color only, sizes and absence of bars on the petals and sepals, all are not recognized here. The species has

an interesting flowering habit; the flowers last two or three weeks on the plant, and opening one at a time. It starts flowering usually in November, and is in full display during December to January. It is not unusual to find the plant in flower during February up to July. The species is named in honor of M. Lüddemann, of Paris.

PHALAENOPSIS MARIAE Burb. Plate 1, fig. 7; Plate 4, figs. 10–18.

Phalaenopsis Mariae BURB. in Warner & Will. Orch. Alb. 2 (1883) *t. 80* et sub. *t. 87;* ROLFE in Gard. Chron. II 26 (1886) 277; HOOK. f. in Bot. Mag. 113 (1887) *t. 6964;* VEITCH, Man. Orch. Pl. pt. 7 (1891) 32; RIDL. in Jour. Linn. Soc. 31 (1896) 292; AMES in Phil. Jour. Sci. 8 (1913) Bot. 434, Orch. 5 (1915) 217, ex Merr. in Jour. Roy. Asiat. Soc. Straits Branch, Special No. (1921) 197, ex Merr. Enum. Phil. Fl. Pl. 1 (1925) 415.

Phalaenopsis Mariae Burb. var. *alba* AMES & QUIS. in Phil. Jour. Sci. 56 (1935) 461, *plate 2, figs. 3 & 4; plate 4, figs. 9 to 17; plate 7, fig. 2.*

The original description is as follows:

Phalaenopsis Mariae. Epiphytal. *Plant* stemless, with flat aërial clinging roots. *Leaves* deflexed, distichous, oblong or ligulate, acute, somewhat channelled, two inches or more in width, stoutish in texture, dark green, glossy, obscurely striate. *Scape* radical, bearing a many-flowered drooping raceme, shorter than the leaves, and proceeding from their axils. *Flowers* of medium size, elegantly coloured; *sepals* narrowly-oblong, bluntish, about an inch long, the lateral ones slightly falcate, white, with about six bold transverse bars or blotches of a deep chocolate red, the basal spots magenta-coloured like the lip; *petals* shorter, broader and more obovate, marked in a similar manner, but with fewer blotches, the colour being the same as in the sepals; *lip* obovate oblong, apiculate, convex, somewhat constricted at the sides, of a rich deep magenta-rose, the middle lobe plane not pilose. *Column* short, white, without fringes at the apex.

—BURB. in Warner & Will. Orch. Alb. 2 (1883) *t. 80* et sub. *t. 87.*

Phalaenopsis (*Stauroglottis*) *Mariae;* caule brevissimo, foliis oblongis v. late lineari-oblongis apicibus acutis saepe recurvis basi uno latere auriculatis, panicula gracili longe pedunculata pluriflora, floribus 1½ poll. latis, sepalis petalisque subaequalibus lineari-oblongis obtusis albis violaceo-fasciatis, labelli lobis lateralibus angustis corniformibus subrecurvis magnibus inflexis, intermedio oblongo purpureo albo marginato basi 2-calcarato, disco villis erectis onuto, columna medio constricta, apice nuda.

—HOOK. F. in Bot. Mag. 113 (1887) *t. 6964.*

Resembles *P. Lueddemanniana* in habit. Leaves linear oblong-oblanceolate, acute, 19 to 40 cm long, 4 to 7 cm wide, dark green, shining above. Scape sparingly branched, few-flowered, 13 to 50 cm long; peduncles and rachis slender, 2 to 2.5 mm

in diameter. Flowers odorless, sometimes slightly fragrant, 2.8 to 3 cm across. Pedicellate ovary slender, white, 1.2 to 1.5 cm long. Lateral sepals obliquely elliptic-ovate, obtuse, apiculate, 1.5 to 1.7 cm long, 0.8 to 0.9 cm wide. Dorsal sepal narrowly oblong, obtuse, 1.4 to 1.7 cm long, 0.7 to 0.9 cm wide. Petals elliptic, obtuse, 1.3 to 1.6 cm long, 6.5 to 8 mm wide. Labellum fleshy, 3-lobed; lateral lobes obliquely oblong, erect, incurved towards the column, 5 to 6 mm long, white, purple and retuse at the apex and base; middle lobe obovate, broad at the apex, 8 to 12 mm long, 6.5 to 8 mm wide at the widest portion, prominently keeled in the middle longitudinally, the keel clothed with hairs on the anterior part, phlox purple except the margins and hairs. Column white, 7 to 8 mm long. Anther cap broadly ovate. Pollinia ellipsoid.

MINDANAO, Lanao Province, Camp Keithley, *Clemens 626, Clemens, s. n.:* Davao Province, Davao, *Loher 6011:* Bukidnon Province, without locality, *Bur. Sci. 21433 Escritor, Bur. Sci. 84781 Quisumbing* (cultivated in Manila); Mt. Dalirig, *Bur. Sci. 21389 Escritor:* without province or locality, *Bur. Sci. 5655 Mrs. Lyons* (cultivated in Manila). In addition to above I have flowers in liquid from plants collected in Cotabato Province and from Jolo. The two collections from Dupax, Nueva Vizcaya Province, Luzon, made by McGregor, previously identified as *P. Mariae,* belong to a form of *P. Lueddemanniana.*

This species is closely allied to *P. Lueddemanniana* Reichb. f. from which it differs in the size of the flowers and in the obtuse sepals and petals. While the typical labellum of *P. Lueddemanniana* has oblong middle lobe, in this species it is obovate, with the apex much broader. The sepals are chartreuse yellow with 4 or 5 chestnut transverse bars. The plant blooms during June to September, usually in July and August. A white variety was reported by Ames and Quisumbing, and this differs from the species in its flowers (pure white except the yellow tips of the sepals and petals). It is known locally as "Flor de la mañana" because of its habit in blooming early in the morning. The species is dedicated to Mrs. Burbidge.

PHALAENOPSIS MICHOLITZII Rolfe. Plate 1, fig. 8; Plate 4, figs. 19–26.

Phalaenopsis Micholitzii ROLFE in Gard. Chron. III 8 (1890) 197, in Journ. des Orch. 1 (1890) 198, in Orch. Rev. 13 (1905) 229; AMES, Orch. 5 (1915) 217, ex Merr. Enum. Phil. Fl. Pl. 1 (1925) 415; AMES & QUIS. in Phil. Jour. Sci. 52 (1933) 454–456, *plate 2, figs. 1 and 2; plate 5, figs. 25 to 33; plate 12, fig. 2.*

The original description is as follows:

From Messrs. F. Sander & Co., St. Albans, came a group of orchids, embracing some fine forms of Cattleya Gaskelliana, C. Dowiana, C. Nilssoni, and C. Schofieldiana; also Masdevallia Amesiana (Veitchi x Tovarense), apricot colour; angraecum articulatum, pure white, the flowers about 1 inch across; and *Phalaenopsis Micholitzii*, the flower of which is greenish white, the long and rather narrow lip white, with coarse hairs and a yellow crest; the leaves are ovate, and shiny-green, about 7 inches in length.—ROLFE, loc. cit. 187.

Herba *P. Lueddemannianæ* habitu. Caulis abbreviatus, paucifoliatus. Folia oblongo-oblanceolata, ad basim sensim angustata, carnosa. Scapi breves, simplices, pauciflori. Flores subflavidi et sine maculis. Sepala lateralia oblique ovata, acuta. Sepalum dorsale oblongo-ellipticum, obtusum. Petala ovato-elliptica, breviter unguiculata. Labellum trilobatum; lobi laterales erecti, subquadrato-oblongi, apice bidentato truncato; lobus intermedius rhombico-spathulatus, inferne unguiculatus, apice obtuse tridentatus; discus supra medium papillis capilliformibus numerosis ornatus. Columna flavida.

Habit similar to that of *P. Lueddemanniana* Reichb. f. Stem abbreviated. Leaves oblong-oblanceolate, 13 to 17.5 cm long, 5.5 to 7 cm wide, broadly obtuse at the apex, gradually tapering to the base, pale green, fleshy, thick, very slightly rigid, somewhat conspicuously nerved with yellowish nerves. Scapes simple, short, few-flowered, 3 to 6 cm long, appearing in the axils of the leaves or at the base of the stem near the roots; rachis very short. Flowers odorless, 6 to 6.5 cm across, yellowish, and absolutely without transverse bars on the sepals and petals, 1 or 2 opening at a time. Pedicellate ovary marguerite yellow, about 3.3 cm long, the ovary terete, not twisted. Lateral sepals obliquely ovate, acute, apiculate, 3.2 to 3.3 cm long, 1.6 to 1.7 cm wide, 9-nerved. Dorsal sepal oblong-elliptic, obtuse, 3.2 to 3.3 cm long, 1.5 to 1.6 cm wide, 9-nerved. Petals ovate-elliptic, obtuse, about 2.8 cm long, 1.7 cm wide, with shortly stalked base which is about 4 mm long, 7-nerved. Labellum fleshy, 3-lobed; lateral lobes erect, subquadrate-oblong, with a prominent fleshy callus above the middle, bidentate at the truncate apex, about 8 mm long, cadmium yellow; middle lobe rhombic-spathulate, about 1.9 cm long, narrowed below into a distinct claw about 7 mm long, obtusely tridentate at the apex when spread out, the irregular margins minutely crisped-undulate, marguerite yellow; disc (between the side lobes) with a ligulate sharply bidentate callus which extends (in the middle of the claw) into a median high keel dentate in front, and which is succeeded by an irregular longitudinal cluster of hair-

like papillæ. Column about 1.2 cm long, marguerite yellow; anther white.

LUZON, Manila, Bureau of Science orchid house, *Bur. Sci. 85572 Eduardo Quisumbing,* February 3, 1932.

A living plant of this species was sent to the author by Mr. F. E. Shafer, an orchid enthusiast of Cebu, who purchased it from a peddler in Cebu. Its origin is unknown, but is doubtless Philippines.

A species with the habit of *P. Lueddemanniana* Reichb. f., differing conspicuously in its yellowish flowers with absolutely no bars on the sepals and petals, and in the rhombic-spatulate middle lobe of the labellum.

EXCLUDED SPECIES

Phalaenopsis cornu-cervi Blume apud NAVES, Novis. App. (1882) 243.
Phalaenopsis deliciosa Reichb. f. apud NAVES, Novis. App. (1882) 243.
Phalaenopsis Devriesiana Reichb. f. apud NAVES, Novis. App. (1882) 243.
Phalaenopsis hebe Reichb. f. apud NAVES, Novis. App. (1882) 242.
Phalaenopsis Lowii Reichb. f. apud NAVES, Novis. App. (1882) 243.
Phalaenopsis Parishii Reichb. f. apud NAVES, Novis. App. (1882) 243.
Phalaenopsis sumatrana Korth apud NAVES, Novis. App. (1882) 242.
Phalaenopsis violacea Teijsm. & Binn. apud NAVES, Novis. App. (1882) 243.

DOUBTFUL SPECIES

PHALAENOPSIS FASCIATA Reichb. f.

Phalaenopsis fasciata REICHB. f. in Gard. Chron. n. s. 18 (1882) 134; ROLFE in Orch. Rev. 13 (1905) 225; AMES, Orch. 5 (1915) 217, ex Merr. Enum. Phil. Fl. Pl. 1 (1925) 414.

The original description is as follows:

This is like *Phalaenopsis sumatrana* in the shape of the light yellow sepals and petals, which have numerous cinnamon bars. The lip has sulphur-colour lateral divisions, which are retuse, and have a blunt keel with a knob parallel to the anterior margin. Between both on the disc is a number of retrorse toothletted orange plates, and two conical papulæ terminating in bristles stand before the base of the median partition. The latter is oblong ligulate (blunt), with a deep, abrupt, membranous keel. The anterior part of it is light purple, the superior orange. There is no cushion of hairs, as in *P. sumatrana* and *Lüddemanniana;* hence, according to artificial characters, it might be regarded as nearest to *Phalaenopsis violacea,* yet the shape of the sepals and petals is markedly different. The sepals have no median keels outside. The top of the lip is totally distinct also. Leaves and roots are said to be quite like those of *Phalaenopsis Lüddemanniana.*

As it is, we cannot now but regard it as distinct, though quite prepared to have one day a rebuke by the occurrence of some intermediate type.

—H. G. Rchb. f.

Phalaenopsis fasciata, n. sp.—Sepals tepalisque oblongis obtusis; labelli partitionibus lateralibus divaricatis retusis cum apiculo latere antico callosis, partitione mediana oblongo-ligulata apice obtusiuscule acuta, lamellis in cristulas solutis in basi; lamelli compresso-conicis aristatis in basi, partitionis anticae carina a basi partitionis madianae in discum, ibi abruptas; columna basi utrinque dilatata. Barba in labelli apice nulla. Folia et radices Phalaenopsidis Lüddemannianae. Sepala ac tepala sulphurea striis cinnamomeis. Labelli partitiones laterales sulphyreae punctulis pallidis cinnamomeis paucis. Partitio mediana postice aurantiaca, antice pallide violaceo-purpurea. Columna basi utrinque purpurea.—Ex Philipp. insul. Imp. cl. Low. H. G. Rchb. f.—Reichb. f. loc. cit. 134.

No material of this species has been seen. Reichenbach f. gave the origin of this plant as Philippines, imported by Messrs. Hugh Low and Co. Reichenbach f. further states that the species is near *P. Lueddemanniana.* Judging by the color of the flower and the description of the flower parts, the species belongs to the *Boxallii* group, *P. Lueddemanniana* differing in the absence of hairs on the crest of the keel of the middle of the lip. The absence of these hairs cannot be used as distinctive and specific character, as this feature is very variable in *P. Lueddemanniana.* A critical examination of the type, if existing, may prove it to be a mere variant of *P. Lueddemanniana,* which is a very variable species.

Phalaenopsis fuscata Reichb. f.

Phalaenopsis fuscata Reichb. f. in Gard. Chron. II 2 (1874) 6; Rolfe in Orch. Rev. 13 (1905) 226; Ames, Orch. 5 (1915) 216, ex. Merr. Enum. Phil. Fl. Pl. 1 (1925) 414.

Phalaenopsis denisiana Cogn. in Gard. Chron. III 26 (1899) 82; Cogn. in Dict. Icon. Orch. (1899) Phalaenop. *t. 6.*

The original description is as follows:

Once more a few Phaloenopsis—now-adays a very unusual source of gratification. It appears to have very large leaves, and I suppose that the inflorescence may be like that of *P. cornu-cervi,* since the plant was well compared with it. The flowers are yellowish, mottled with brown, and very fleshy. The lip is quite peculiar, and the lateral sepals are not so much extended as in *P. cornu-cervi.* I have to thank for it Mr. Bull, who introduced it from the Malay Peninsula.—H. G. Rchb. f.

Aff. *P. cornu-cervi,* radicibus brevibus; foliis amphissimis oblongis obtuse acutis (pedunculo certe *P. cornu-cervi?*); floribus mediocribus illos speciei dictae acquantibus; sepalis oblongis obtuse acutis; tepalis cuneato-oblongis obtusis; labello tripartito, partitionibus lateralibus ligulatis retusis utrinque

unidentatis, latere inferiore medio umbonatis, partitione media oblonga acuta, per medium carinata; callo bidentato in basi, postposita ligual aristata utrinque, columna basi exangulata.—REICHB. F. loc. cit. 6.

The origin of *P. fuscata* was reported as the Malay Peninsula; that of *P. denisiana* as Philippines. I have on hand material of so called *P. fuscata,* an imported plant from Singapore. If my material is indeed a *fuscata,* it is distinct, and is closely allied to *P. Lueddemanniana.* No material of *P. denisiana* has been seen.

PHALAENOPSIS PALLENS (Lindl.) Reichb. f.

Phalaenopsis pallens (Lindl.) REICHB. f. in Walp. Ann. 6 (1864) 932; ROLFE in Gard. Chron. II 26 (1886) 276, in Orch. Rev. 8 (1900) 327, 13 (1905) 226.

Trichoglottis pallens LINDL. in Jour. Hort. Soc. 5 (1850) 34, in Paxt. Flow. Gard. 1 (1850) 15.

Stauropsis pallens REICHB. f. in Hamb. Gartenz. 16 (1860) 117, Xen. Orch. 2 (1862) 7; NAVES, Novis. App. (1882) 243.

For many years this species was ascribed to the Philippines. It does not occur in the Archipelago, and Rolfe, loc. cit., has shown that the type could not have come from the Philippines.

PHALAENOPSIS REICHENBACHIANA Reichb. f. and Sander.

Phalaenopsis Reichenbachiana REICHB. f. & SANDER in Gard. Chron. II 18 (1882) 586; ROLFE in Orch. Rev. 13 (1905) 226; AMES, Orch. 5 (1915) 218, ex Merr. Enum. Phil. Fl. Pl. 1 (1925) 416.

No material of this species has been seen. According to Rolfe (Orch. Rev. loc. cit.) Micholitz stated that this species is a native of Mindanao. By its description it is perhaps a *P. Lueddemanniana.*

PHALAENOPSIS VEITCHIANA Reichb. f.

Phalaenopsis Veitchiana REICHB. f. in Gard. Chron. (1872) 935; BURB. in Floral Mag. 15 (1876) *t. 213;* VEITCH, Man. Orch. Pl. pt. 7 (1898) 47; AMES, Orch. 5 (1915) 218, ex Merr. Enum. Phil. Fl. Pl. 1 (1925) 417; G. WILSON in Orch. Rev. 30 (1922) 346.

Rolfe [11] suggested that this species is a hybrid between *P. Schilleriana* and *P. equestris,* and mentioned the fact the middle lobe of the lip has anchorlike appendages. An examination of the type, which I have not seen, will throw light of its status and its relation to *P. Gertrudae,* which is a natural hybrid between *P. equestris* and *P. Schilleriana.*

[11] See Ames in Phil. Jour. Sci. 4 (1909) Bot. 599.

ILLUSTRATIONS

[The colored drawings were made by Mr. Pedro L. Ramos and the line drawings by Mr. Ricardo C. Aguilar, both scientific illustrators of the Natural History Museum]

PLATE 1

FIG. 1. *Phalaenopsis equestris* (Schauer) Reichb. f. Front view of flower, × 1.

2. *Phalaenopsis Lindenii* Loher. Front veiw of flower, × 1.
3. *Phalaenopsis Lueddemanniana* Reichb. f. Front view of typical flower, × 1.
4. *Phalaenopsis Lueddemanniana* Reichb. f. Side view of flower, the form with greenish background, × 1.
5. *Phalaenopsis Lueddemanniana* Reichb. f. Front view of flower, the *Boxallii* form with yellow background and ferruginous bars, × 1.
6. *Phalaenopsis Lueddmanniana* Reichb. f. Side view of lip, × 2.
7. *Phalaenopsis Mariae* Burb. Front view of flower, × 1.
8. *Phalaenopsis Micholitzii* Rolfe. Front view of flower, × 1.

PLATE 2

Phalaenopsis equestris (Schauer) Reichb. f.: *1*, habit of the plant, one-third natural size; *2*, from view of flower, × 1; *3*, side view of flower, × 1; *4*, dorsal sepal, × 2; *5*, petal, × 2; *6*, lateral sepal, × 2; *7*, side view of column, × 2; *8*, front view of column, × 2; *9*, labellum from above (stretched out), × 2; *10*, anther cap, from above, × 5; *11*, anther cap from below, × 5; *12*, pollinia, × 5.

PLATE 3

Phalaenopsis Lueddemanniana Reichb. f.: *1*, habit of plant, × 0.5; *2*, dorsal sepal, × 1; *3*, lateral sepal, × 1; *4*, petal, × 1; *5*, one form of labellum (expanded), × 2; *6*, another form of labellum (expanded), × 2; *7*, still another form of labellum (expanded), × 2; *8*, side view of column and labellum, × 2; *9*, front view of column and labellum, × 2; *10*, anther cap from below, × 5; *11*, anther cap from above, × 5; *12*, pollinia, × 5.

PLATE 4

Phalaenopsis Lindenii Loher: *1*, dorsal sepal, × 2; *2*, lateral sepal, × 2; *3*, petal, × 2; *4*, labellum (expanded), × 2; *5*, front view of column, × 2; *6*, side view of column, × 2; *7*, anther cap from above, × 5; *8*, anther cap from below, × 5; *9*, pollinia, × 10.

Phalaenopsis Mariae Burb.: *10*, dorsal sepal, × 2; *11*, lateral sepal, × 2; *12*, petal, × 2; *13*, front view of column and labellum, × 2; *14*, labellum (expanded), × 2; *15*, side view of column and labellum, × 2; *16*, anther cap from above, × 5; *17*, anther cap from below, × 5; *18*, pollinia, × 10.

Phalaenopsis Micholitzii Rolfe; *19*, dorsal sepal, × 1; *20*, lateral sepal, × 1; *21*, petal, × 1; *22*, labellum (expanded), × 2; *23*, side view of column and labellum, × 2; *24*, front view of column and labellum, × 2; *25*, anther cap from above, × 5; *26*, pollinia, × 5.

PLATE 5. PHALAENOPSIS LINDENII LOHER

FIG. 1. Habit with leaves and flowers, much reduced.

2. Portion of leaf showing maculations and tip of inflorescence with buds and opened flower, slightly enlarged.

PLATE 1.

PLATE 2.

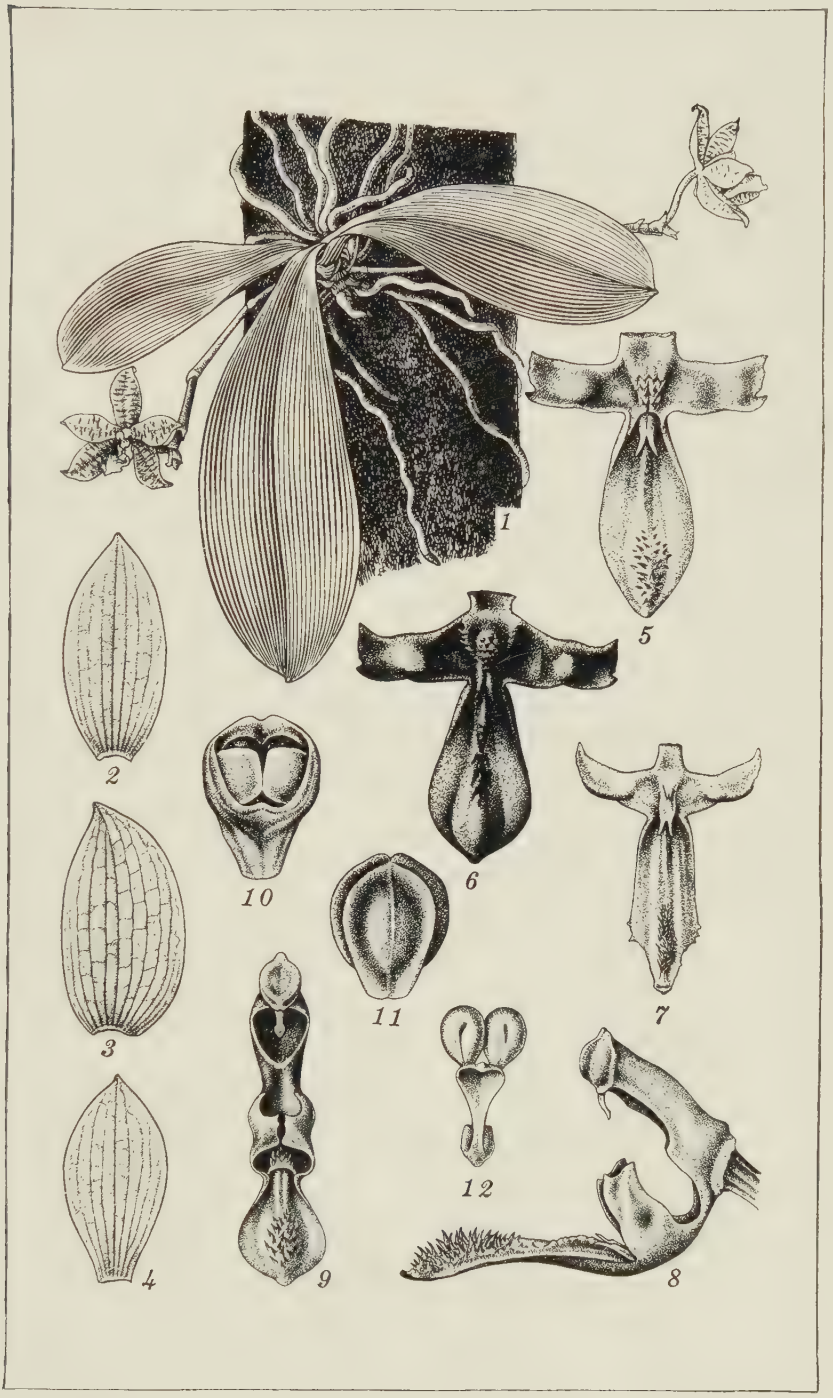

PLATE 3.

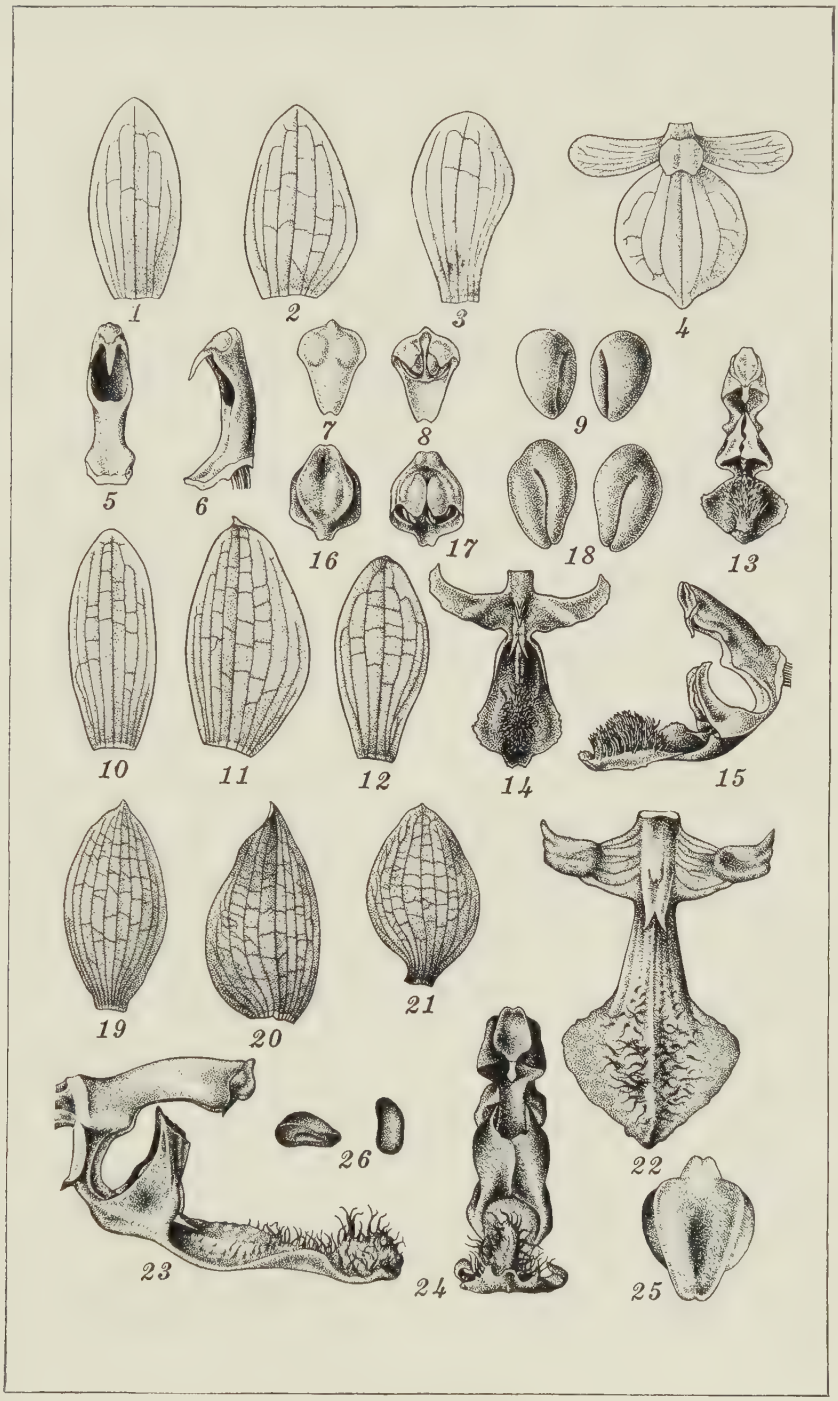

PLATE 4.

PLATE 5.

NOTES ON THE ANTHELMINTIC PROPERTIES OF THE LATEX OF PAPAYA (CARICA PAPAYA LINN.) AND OF "ISIS" (FICUS ULMIFOLIA LAM.)

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According to Tavera (1892), Guerrero (1921), and other botanical writers, there are many species of plants in the Philippines which are of medical importance. Some of these plants are of known therapeutic value and appear in contemporary pharmacopoeias, according to Valenzuela, Concha, and Santos (1946). There are others, however, the efficacy of which has not yet been accurately determined.

The purpose of this paper is to record the results of a study on the anthelmintic properties of a few common plants. The latex of the following nine species representing three families was examined: (1) Moraceæ—*Ficus balete* Merr., *F. nota* (Blanco), *F. odorata* (Blanco), *F. pisifera* Wall., *F. ulmifolia* Lam., *Castilloa elastica* Cerv., and *Artocarpus integra* Thunb.; (2) Sapotaceæ—*Achras zapota* Linn.; and (3) Caricaceæ—*Carica papaya* Linn. Several members of the genus *Ficus* were included in the study because of their systematic relationship with *Ficus doliaria,* a South American wild fig, the latex of which has been proven to be an efficient anthelmintic against ascarids and trichurids. In the case of papaya, according to Tavera (1892) and Berger and Asenjo (1940), the crude latex has long been known to have anthelmintic properties, but the available literature does not show that its efficacy has been critically tested.

METHODS

Collection and preservation of latex.—Latex samples were obtained by wounding the trunk, stems, and unripe fruits of a plant with a clean knife and placing the partly coagulated milky juice that exudes in a bottle containing sodium benzoate dissolved in normal salt solution. The proportion of latex to salt solution was 4 to 1 and the final concentration of the sodium benzoate 1 per cent. The samples were kept at room temperature and used within one week after collection. Some

samples were mixed with two to three volumes of alcohol and the precipitated proteinates were filtered off, dried over calcium chloride, and ground into coarse powders.

In vitro tests.—The samples were screened by means of the worm-digesting method of Robbins (1930). One or two live *Ascaris lumbricoides* collected from swine were immersed in a 5 per cent emulsion of latex, or 1 per cent emulsion of proteinate derivative, in Ringer's solution. Another set of worms immersed in Ringer's solution alone served as control. The parasites were then placed in an incubator at 37° C. and examined at one-hour intervals for any evidence of anthelmintic effect.

In vivo tests.—The samples that showed marked anthelmintic activity *in vitro* were selected for further study. These samples were tested for toxicity by feeding them in large doses to guinea pigs and rats. If found nontoxic, they were given in varying amounts to young dogs and human volunteers infected with different kinds of intestinal worms. They were mixed with two volumes of water and a little amount of sugar and given early in the morning on an empty stomach followed after one or two hours with sodium sulphate. The human cases were worm-egg-counted before and two to three weeks after treatment. The dogs were worm-egg-counted before treatment and on the third day after treatment they were sacrificed and examined for parasites. The fæces of all the cases passed during the first twenty-four hours after treatment were collected and sieved for the presence of worms.

RESULTS

In Table 1 are summarized the results of the *in vitro* tests. Of the nine species of plants tested only *Carica papaya* and *Ficus ulmifolia* were found to possess marked anthelmintic properties. The others were either inert or only slightly active. The *Ascaris* worms placed in the latex of *Carica papaya* and of *Ficus ulmifolia* were either dead or moribund one hour after immersion, and their cuticles showed the presence of small blisters in several places. Some of these blisters eventually ruptured, allowing the reproductive organs of the parasites to protrude through the openings. The worms appeared much distorted, later undergoing more or less complete disintegration. Worms placed in 1 per cent emulsions of the proteinate deri-

vatives prepared from the saps of the two plants were similarly affected.

TABLE 1.—*In vitro effect of the latex of plants on Ascaris lumbricoides.*

Kind of plant	Effect after—			
	1 hour	2 hours	4 hours	8 hours
Ficus balete	Alive	Alive	Alive	Alive.
Ficus nota	do	do	do	Do.
Ficus odorata	do	do	Moribund	Dead, with few blisters.
Ficus pisifera	do	do	Alive	Alive.
Ficus ulmifolia	Dead, with few blisters	Ulcerated	Ulcerated	Body much distorted.
Artocarpus integra	Alive	Alive	Alive	Alive.
Castilloa elastica	do	do	do	Do.
Achras zapota	do	do	do	Do.
Carica papaya	Moribund	Dead, with blisters	Ulcerated	Body much distorted.
Control: Ringer's solution.	Alive	Alive	Alive	Alive.

The results of the treatment are shown in Tables 2 and 3. Four pups infected with ascarids (*Toxocara canis*) and hookworms (*Ancylostoma caninum*) were given 5 mils each of papaya latex. Twenty-eight dead ascarids were collected from the fæces of these animals on the first day of treatment, but no hookworms were found. At autopsy large numbers of hookworms were recovered from the intestines of each, but all of them were free of ascarids. The efficiency of papaya latex in this series of animals is thus 100 per cent against ascaris and apparently 0 per cent against hookworms.[1]

Four persons infected with *Ascaris* and *Trichuris* were given papaya latex in doses of 30 to 50 mils depending upon age and size. All of them passed dead worms during the first day of treatment, but when examined two weeks later one was still positive for *Ascaris* and three still harbored *Trichuris* (Table 3). There was, however, a 44.4 per cent reduction in the *Ascaris* egg count of the person still positive for *Ascaris* and an average of 58.5 per cent reduction in the *Trichuris* egg

[1] In later experiments it was determined that the latex of *Carica papaya* and of *Ficus ulmifolia* has no effect *in vitro* on live dog hookworms.

counts of the three still positive for *Trichuris*. The efficiency of papaya latex in this series is thus 79.6 per cent against *Ascaris* and 71 per cent against *Trichuris*.

TABLE 2.—*Effect of papaya latex on Toxocara canis in dogs.*

Dog Number	Weight	Dose	Worms recovered from faeces	Worms found at autopsy	Reduction
	kg.	*ml.*			*Per cent*
1	1.2	5.0	6	0	100
2	1.5	5.0	12	0	100
3	1.4	5.0	3	0	100
4	1.5	5.0	7	0	100
Total			28	0	100

TABLE 3.—*Effect of the latex of Carica papaya and of Ficus ulmifolia on intestinal worms in man.*

Name	Age	Sex	Dose	Egg counts per ml. of faeces						Worms recovered from faeces
				Before treatment			After treatment			
				Ascaris	*Trichuris*	Hookworms	*Ascaris*	*Trichuris*	Hookworms	
	Years		*ml.*	*Carica papaya series*						
L. N.	15	F	40	6,500	600	-------	-------	-------	-------	4 *Ascaris*.
R. R.	10	M	30	20,500	2,900	-------	11,400	1,900	-------	2 *Ascaris*, 2 *Trichuris*, 4 pinworms.
E. R.	12	F	30	12,000	3,100	-------	-------	400	-------	3 *Ascaris*, 5 *Trichuris*, 4 pinworms.
B. H.	54	M	50	17,000	1,000	-------	-------	600	-------	----------
				Ficus ulmifolia series						
A. N.	13	F	15	70,000	2,500	1,200	-------	150	1,400	21 *Ascaris*, 4 *Trichuris*.
D. M.	24	M	25	12,500	3,600	-------	-------	-------	-------	8 *Ascaris*, 6 *Trichuris*.
S. A.	46	M	30	-------	5,600	-------	-------	600	-------	14 *Trichuris*, 12 pinworms.

Three persons were given *Ficus ulmifolia* latex in doses of 15 to 30 mils each. They all passed dead worms during the first day of treatment. The two cases infected with *Ascaris* were found to be free of the parasite when examined three weeks later. Of the three individuals infected with *Trichuris* only one was completely cured, but there was an average reduction of 91 per cent in the *Trichuris* egg counts of the other two. There was no significant change in the hookworm egg counts of the individual infected with hookworms before and after the treatment. The efficiency of the latex of *Ficus ulmi-*

folia in this small series is thus 100 per cent against *Ascaris*, 93.6 per cent against *Trichuris*, and 0 per cent against hookworms.

Two persons in the papaya group and one in the *Ficus* group passed some pinworms (*Enterobius vermicularis*) along with other dead parasites, indicating that the saps of *Carica papaya* and *Ficus ulmifolia* also have enterobicidal properties.

The ascarids recovered from the fæces of the dogs and the human cases showed blisters and ulcers on their cuticles, and some were broken into fragments and in advanced stages of degeneration. A few *Trichuris* were also blistered, but their bodies were intact. The pinworms did not appear damaged externally.

DISCUSSION

The results of the various tests show that the anthelmintic properties of the saps of *Carica papaya* and *Ficus ulmifolia* are similar to those of higuerolatex, as reported by Caldwell and Caldwell (1929), Brooks and Brown (1942), and others. The latex of *Ficus ulmifolia* appears to be more efficient than papaya latex, but unfortunately it is difficult to obtain in large quantities. Both products were well tolerated by the cases treated, but one contraindication against their use is the presence of open lesions in the digestive tract. This is due to the fact that the effective anthelmintic principles are proteolytic enzymes (ficin and papain) which are capable of digesting not only live worms but also injured mucous membranes.

SUMMARY

The latex of *Carica papaya* and of *Ficus ulmifolia* out of nine species of plants tested was found to possess anthelmintic properties against ascarids, trichurids, and pinworms. Papaya latex was 100 per cent effective against the dog ascarid, 79.6 per cent against human *Ascaris* and 71 per cent against *Trichuris*. The latex of *Ficus ulmifolia* was 100 per cent against *Ascaris* and 93.6 per cent against *Trichuris*. Both products were inactive against hookworms.

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THE TREATMENT OF FASCIOLIASIS IN DAIRY CATTLE AND IN INDIAN BUFFALOES WITH HEXACHLORETHANE AND KAMALA EXTRACT

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Fascioliasis, or liver rot, is one of the most destructive of the parasitic diseases of ruminants in the Philippines. It is caused by either one or both of *Fasciola hepatica* Linn. and *F. gigantica* Cobbold which infect upwards from 1.66 to 19 per cent of cattle and/or carabaos, *Bubalus bubalis* Ledg. (Robles, 1932; De Jesus, 1938; Arañez, unpublished). Alone, this scourge has been responsible for the condemnation of no small number of liver portions or even of the whole organ, apart from the considerable loss caused by retarded growth, lowered milk production, curtailed breeding activity, emaciation, and death of infected animals. Thus, it is an economic problem of great concern both to the stockman and to the veterinarian.

Owing to the above considerations, and in keeping with the general program of this institution of finding cheap expedients (and where known, to determine their relative efficacy) for the treatment of the more important parasites of livestock, hexachlorethane-kamala extract mixture was tried against this infection in dairy cattle and in Indian buffaloes, *Bubalus buffelus*.

REVIEW OF THE LITERATURE

Although the discovery by Jehan de Brie of *Fasciola hepatica* as the causative agent of sheep liver rot was made as early as 1379, it was in the nineteenth century that the treatment for this disease really gained impetus and has since engaged the attention of various workers throughout the world. Grassi and Calandruccio (1884) appear to have pioneered in the medication of this scourge in sheep using extract of male fern. Giving orally a single dose of 5 grams of ethereal extract of male fern in 50 grams of the ethereal tincture, these workers observed the expulsion of numerous flukes in the feces after 24 to 48 hours and the disappearance after the third day of the eggs in the dung and of the adult worms at autopsy. Two years later (1886) Perroncito tried the same experiment.

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While he got marked reduction in the quantity of eggs in the dejecta, he likewise obtained some unfavorable effects on the host particularly severe flatulence which, fortunately, subsided in about an hour. Alessandrini (1908), however, observed differently. Using also extract of male fern in two severely infected sheep, he got a disheartening result—the death of both parasites and hosts. In the same species of animal Railliet, Moussu, and Henry (1911) used 5 grams of the ethereal extract in 25 cc of oil given in from 1 to 4 doses on successive days. Finding it effective, they suggested its use at the dose rate of 1 gram of the extract per 5 kilos of body weight. Montgomerie (1925) found oleoresin of aspidium in milk an efficient flukecide for the adult worms, but is rather ineffectual for the immature parasites.

In cattle Borini (1911) tried the ethereal extract of male fern consistently getting favorable results in light infections but not in heavily infected cases with cachexia.

After these early experiments, a number of proprietary products of male fern appeared in many European markets under the trade names of "distol" (manufactured in Hungary), "danistol" (believed to be similar to distol), "fasciolin," "avisciolina," "filmaron," etc. Distol was recommended by Marek (1917) and by Kraneveld (1925). Only lately Swanson and Goo (1938), Alicata, et al. (1940), and Alicata (1941) found it effective against fascioliasis in cattle, but the milk acquired a bitter salty taste that lasted for a few days. Danistol is much more expensive and yet no more effective than distol, according to Montgomerie (1926).

Other nonmale fern preparations had also been tried, like calomel, sodium salicylate, compounds of arsenic, phosphorus, mercury and antimony, tetrachlorethylene, carbon tetrachloride, kamala, hexachlorethane alone and the latter's combination with tetrachlorethylene, filicic acid, kamala extract, and inert ingredients, but, save for the last seven, all had been found ineffective. Carbon tetrachloride which gave satisfactory results to Ernst (cited by Chopra and Chandler, 1928) and to Montgomerie (1926) in sheep was considered by Hutyra and his associates (1938) and by Monnig (1938) to be dangerous for ruminants and rather toxic for cattle, producing central necrosis and fatty degeneration of the liver especially among fattened animals and those with hypocalcemia, in advanced pregnancy, and in lactation. Kamala, while effective, was

observed by Alicata, et al. (1940) and by Alicata (1941) to produce profuse and weakening diarrhea which lasted for as long as two weeks.

Hexachlorethane alone was well recommended by De Blieck and Baudet (1928) and by Noller (cited by Monnig, 1938) for cattle fascioliasis. While found to be highly efficacious by Hilz and Schauble in doses of 20 to 30 grams per 50 kilos live weight, according to Hall as cited by Alicata (1941), it was observed by Noller and by Alicata to cause colic in milch cows feed on concentrates, or when given in high concentrations. Marek (1926), Thienel (1927), and Alicata (1941) combined this flukecide with tetrachlorethylene, filicic acid, and kamala extract, respectively, while Vianello (1937), Pegreffi (1939), Rosenberger and Slesic (1942), and Olsen (1943, 1944) mixed it with inert ingredients. Olsen used hexachlorethane in aqueous suspension with bentonite as a drench which, although he claimed to have gotten highly encouraging results (91 per cent efficiency) over his one-day treatment for fascioliasis hepatica, was found in Hawaii that the "results with this method have not been very satisfactory" (Alicata in a personal communication to the writer January 12, 1946).

MATERIALS AND METHODS

The subjects for this study were forty-eight dairy cattle (mostly grades) and four Indian buffaloes belonging to the Swiss Dairy Farm at Caloocan, Rizal, Philippines. The concern had formerly about a hundred of these animals but many died of fascioliasis prior to the treatment. Hexachlorethane and kamala extract were given in capsules at the rate of 10 grams and 1.75 grams, respectively, for every 30 kilos of body weight. The total dose was divided into approximately equal quantities and was administered orally over two successive days following an overnight fasting (Table 1). Feed was likewise withheld at least three more hours after each dose. As it was thought that *therapia sterilisans magna* might be possible with a single treatment (for practical purposes), four of the cows were given the total amount only once (Table 2) instead of distributing it over a two-day period, as suggested by Alicata (1941). In two others the total dose was given daily for two consecutive days. Single injections of 20 per cent calcium-borogluconate solution were given the animals the better number of which were poor risks.

TABLE 1.—*Showing the effect on fascioliasis of the total amount of 10 grams hexachlorethane and 1.75 grams kamala extract per 30 kilos body weight equally distributed over a two-day period.*

Animal No.	Weight	Flukecide, first day		Flukecide, second day		Egg-count per gram of feces		Flukecide efficiency against mature flukes	Necropsy findings	Remarks
		Hexachlorethane	Kamala extract	Hexachlorethane	Kamala extract	Pre-treatment	Post-treatment			
	Kilos	*Grams*	*Grams*	*Grams*	*Grams*			*Per cent*		
22	263	43.80	7.66	43.80	7.66	110	22	80.00	Some adult flukes found.[a]	Lively, appetite good throughout. Slight diarrhea noted.
66	309	51.50	9.01	51.50	9.01	88	0	100.00	Negative for flukes; liver appeared normal.	Profuse diarrhea for 3 days. Appetite fair.
38	324	54.00	9.45	54.00	9.45	132	22	83.33	Some flukes found.[a]	Disintegrated flukes in feces after 3 days; no appetite and profuse diarrhea for 2 days.
67	253	42.15	7.37	42.15	7.37	44	0	100.00	Negative.	Disintegrated flukes seen in feces after 4 days. Lively; appetite fair.
27	276	46.00	8.05	46.00	8.05	44	0	100.00	Four immature flukes found.	Fair appetite; lively.
68	306	51.00	8.92	51.00	8.92	198	66	66.66	Many adult flukes found.[a]	Do.
32	277	46.15	8.07	46.15	8.07	220	44	80.00	Some adult flukes found.[a]	Fair appetite; slight diarrhea for 5 days.
57	293	48.65	8.48	48.65	8.48	66	0	100.00	Negative for flukes; liver appeared normal.	Good appetite.
43	288	48.00	8.40	48.00	8.40	22	0	100.00	-----do-----	Do.
84	250	41.60	7.28	41.60	7.28	44	0	100.00	-----do-----	Do.
86	243	40.50	7.08	40.50	7.08	88	22	75.00	Some adult flukes found.[a]	Fair appetite; lively.
72	321	53.50	9.36	53.50	9.36	44	0	100.00	Negative	Good appetite; slight diarrhea.
95	274	45.66	7.98	45.66	7.98	22	0	100.00	-----do-----	Do.

41	247	41.15	7.20	41.15	7.20	66	0	100.00	----do----	Fair appetite; lively; slight diarrhea for 3 days.
30	257	42.80	7.49	42.80	7.49	66	22	66.66	Some adult flukes found.[a]	Good appetite; lively.
60	298	49.65	8.68	49.65	8.68	44	0	100.00	Negative.	Fair appetite; slight diarrhea for 6 days.
89	310	51.50	9.01	51.50	9.01	44	0	100.00	----do----	Profuse diarrhea noted; appetite poor.
83	293	48.80	8.54	48.80	8.54	22	0	100.00	----do----	Poor appetite for 2 days.
81	316	52.65	9.21	52.65	9.21	66	22	66.66	Some adult flukes noted.[a]	Fair appetite on day of treatment.
40	262	43.65	7.63	43.65	7.63	22	0	100.00	Negative.	Good appetite; disintegrated flukes seen in feces after 3 days.
88	241	40.15	7.02	40.15	7.02	22	0	100.00	Three young flukes found.	Fair appetite; lively.
96	339	56.50	9.88	56.50	9.88	22	0	100.00	Negative.	Lively; good appetite.
85	253	42.15	7.37	42.15	7.37	132	44	66.66	Some adult flukes noted.[a]	Good appetite.
92	326	54.30	9.50	54.30	9.50	22	0	100.00	Negative	Fair appetite.
73	293	48.80	8.54	48.80	8.54	44	0	100.00	Negative	Disintegrated flukes seen in stool after 2 days. Lively; good appetite.
28	260	43.30	7.57	43.30	7.57	88	22	75.00	Some adult flukes found.[a]	Good appetite.
24	338	56.30	9.85	56.30	9.85	22	0	100.00	Negative.	Diarrhea for 5 days; appetite poor.
26	259	43.15	7.55	43.15	7.55	22	0	100.00	Negative.	Fair appetite.
11	287	47.80	8.36	47.80	8.36	88	22	75.00	Some adult and immature flukes found.[a]	No appetite for a day; lively; slight diarrhea.
42	289	48.15	8.42	48.15	8.42	44	0	100.00	Negative.	No appetite for 2 days; lively.
45	250	41.60	7.28	41.60	7.28	110	22	80.00	Some adult flukes found.[a]	Good appetite.

TABLE 1.—*Showing the effect on fascioliasis of the total amount of 10 grams hexachlorethane and 1.75 grams kamala extract per 30 kilos body weight equally distributed over a two-day period*—Continued.

Animal No.	Weight	Flukecide, first day		Flukecide, second day		Egg-count per gram of feces		Flukecide efficiency against mature flukes	Necropsy findings	Remarks
		Hexachlorethane	Kamala extract	Hexachlorethane	Kamala extract	Pre-treatment	Post-treatment			
	Kilos	*Grams*	*Grams*	*Grams*	*Grams*			*Per cent*		
87	280	46.65	8.16	46.65	8.16	22	0	100.00	Negative.	Fair appetite; disintegrated flukes in feces seen after 3 days.
69	293	48.80	8.54	48.80	8.54	66	0	100.00	do	Slight diarrhea for 4 days.
44	251	41.80	7.31	41.80	7.31	154	44	71.42	Some adult flukes found.	Good appetite; slight diarrhea for 2 days.
39	247	41.10	7.20	41.10	7.20	110	22	80.00	do	Good appetite; lively.
78	288	48.00	8.40	48.00	8.40	44	0	100.00	Negative.	Profuse diarrhea for 4 days.
63	269	44.80	7.74	44.80	7.74	66	0	100.00	do	Do.
48	254	42.30	7.40	42.30	7.40	22	0	100.00	do	Slight diarrhea for 3 days; good appetite.
25	293	48.80	8.54	48.80	8.54	44	0	100.00	do	Good appetite.
46	301	50.15	8.77	50.15	8.77	44	0	100.00	do	Profuse diarrhea for 2 days.
51	242	40.33	7.05	40.33	7.05	66	0	100.00	do	Profuse diarrhea for 3 days.
14	248	41.30	7.22	41.30	7.22	110	22	80.00	Some adult flukes found.[a]	Lively; good appetite.
Buffalo 1	486	81.00	14.17	81.00	14.77	44	0	100.00	Negative.	Profuse diarrhea for 4 days; lively.
Buffalo 2	482	80.35	14.06	80.35	14.06	220	66	70.00	Some adult and immature flukes found.[a]	Slight diarrhea for 5 days.
Buffalo 3	507	84.50	14.78	84.50	14.78	66	0	100.00	Negative.	Slight diarrhea for 6 days; good appetite.
Buffalo 4	498	83.00	14.52	83.00	14.52	110	22	80.00	Some adult flukes found.	Slight diarrhea for 3 days; good appetite and lively.
Average anthelmintic efficiency								91.22		

[a] Only livers of animals with negative feces were meticulously examined postmortem to verify laboratory findings because a thorough inspection of these organs will result in their devaluation.

TABLE 2.—*Showing the effect on fascioliasis of the total amount of 10 grams kamala extract per 30 kilos body weight given only once or daily for two consecutive days.*

Animal No.	Weight	Flukecide, first day		Flukecide, second day		Egg-count per gram of feces		Flukecide efficiency against mature flukes	Necropsy findings	Remarks
		Hexachlorethane	Kamala extract	Hexachlorethane	Kamala extract	Pre-treatment	Post-treatment			
	Kilos	*Grams*	*Grams*	*Grams*	*Grams*			*Per cent*		
47	268	89.30	15.62	----------	----------	132	0	100.00	Negative for flukes.	Full dose given once, profuse diarrhea for a week; appetite good; lively.
35	262	87.30	15.27	----------	----------	154	----------	----------	All mature flukes disintegrating; liver appeared half-cooked, immature flukes unaffected.	Full dose given once, down and prostrate on the second day after treatment; profuse diarrhea, died two days thereafter.
21	238	79.30	13.87	----------	----------	132	----------	----------	----do----------	Full dose given once, down on fourth day after treatment, profuse diarrhea, died two days thereafter.
19	247	82.30	14.40	----------	----------	110	0	100.00	Negative for flukes.	Full dose given once, profuse diarrhea for 4 days; lively; appetite fair.
36	232	77.30	13.52	77.30	13.52	176	----------	----------	All flukes disintegrating, liver appeared half-cooked.	Emaciated animal; full dose given twice; down on the following day after last dose, died on 3rd day.
23	239	79.60	13.93	79.60	13.93	244	----------	----------	All flukes disintegrating, necrotic areas present, liver appeared half-cooked.	Full dose given twice, down on 3rd day, profuse diarrhea, died 2 days thereafter.

Precautions were taken to preclude the reinfection of the herd during the experiment.

The differential-egg-count test which is commonly employed in the determination of the anthelmintic efficacy of expedients (Moskey and Harwood, 1941), subsequently checked by necropsy findings, was used as the criterion for evaluating the efficiency of the hexachlorethane-kamala extract. Shortly before and a month after treatment, a 200-gram fecal sample was obtained rectally from each ruminant for three consecutive days and the samples were deposited in correspondingly labelled bottles. Those of the same subject were grouped together and after their thorough comminution the ova in each sample were counted, using the dilution-egg-count technic of Whitlock (1941), which is a modification of Gordon's and Whitlock's (1939). Briefly, the method was as follows: A 10-gram stool was placed in a bottle and enough water was added up to the 150-cc level. After thorough stirring, about 10-cc suspension was strained through an 18-mesh wire gauze and 0.5 cc of the latter was drawn into a tuberculin syringe. Saturated salt solution was subsequently drawn in until the contents reached the 1-cc mark. This was followed shortly by the suction of an air bubble with sufficient diameter capable of moving up and down freely when the syringe is lifted. Then an even suspension was secured by tilting the syringe up and down with the air bubble, the contents being agitated considerably. After about 0.2 cc as waste was withdrawn, and before the suspensoids could settle down, three 0.15-cc samples were immediately smeared on three slides. The eggs were now counted, and the average of all the egg-counts in the three smears multiplied by 200 gave the number of ova per gram of dung.

Three such counts were made for every sample collected from each subject prior to the treatment, and the average of all the nine counts was taken as the index of the quantity of eggs per gram of dejecta of that animal. Analogous counts were also made from the collections obtained a month after the medication, and, the difference between the pre- and the post-treatment egg-counts being known, it was then easy to determine the efficiency of the expedient by simple mathematical calculation.

Two months later, and following consultation with the writer who was not averse to the idea, the manager sent all the animals to the block, because he feared that they would only

get lost on account of the disorder then obtaining during the Japanese occupation. To the writer, this act was most welcome, because, aside from saving the concern from augmenting its losses, it also offered him the opportunity to examine the liver, thus enabling him to determine the effect of his treatment.

OBSERVATIONS AND RESULTS

The observations and results are presented in Tables 1 and 2. Table 1 shows the effect on fascioliasis of the total amount of 10 grams hexachlorethane and 1.75 grams kamala extract per 30 kilos body weight equally distributed over a two-day period. Table 2 shows the effect on fascioliasis of the total amount of 10 grams hexachlorethane and 1.75 grams kamala extract per 30 kilos body weight given only once or daily for two consecutive days.

DISCUSSION

The total dose of 10 grams hexachlorethane and 1.75 grams kamala extract per 30 kilos body weight administered over a two-day period was apparently well tolerated by the test subjects (Table 1), but not so with the bigger dosages especially when dealing with debilitated animals (Table 2). Encouraging results were obtained with the former dose, and from forty-six animals parasitized with either one or both of *Fasciola hepatica* and *F. gigantica,* the average anthelmintic efficiency obtained was 91.22 per cent. The efficiency, however, seems to depend on the intensity of infection. Adult worms were conspicuous by their absence in the liver of posted animals having pretreatment counts as high as 44 eggs per gram of dung. This egg level is higher than that observed by Alicata (1941) who found "that in cases where the egg count was below 35 eggs per gram of feces, this dosage completely eliminated all adult flukes, as evidenced by subsequent absence of fluke eggs in the feces." Where the egg count per gram was as high as 66 eggs, the efficiency in eight animals varied from 66.66 to 100 per cent, with an average of 91.66 per cent. The average in four cases with 88 eggs per gram of dejecta was 81.25 per cent, and 80 per cent in five cases where the count per gram was as high as 110 ova. Due to the paucity of data, no mention could be made of the cases with counts beyond 110 eggs per gram of stool.

Adult flukes undergoing degeneration were found in the feces of animals treated 2 to 4 days previously. Young flukes seem

not to be affected by the expedient for, with reinfection forestalled, worms short of gravidity were still seen in the livers of cows number 27, 88, and 11 and from the same organ of buffalo number 2 killed two months after deworming. Furthermore, live flukes in a much younger state of development than the preceding were encountered along with adult parasites that were undergoing disintegration in one of the animals (cow number 35) that died on the fourth day following the administration of a big dose (Table 2).

It may be recalled that Olsen in 1941 stated that he obtained 91 per cent efficiency over his one-day treatment using hexachlorethane in aqueous suspension with bentonite as a drench (*vide supra*), adding that "treatments of cattle with hexachlorethane alone, or hexachlorethane and kamala, in capsules, did not give results superior to the drench method." On the other hand, Alicata [2] in a personal communication to the writer mentioned that results obtained with the Olsen's method "have not been very satisfactory." Results obtained by the writer with hexachlorethane-kamala extract in capsules against fascioliasis hepatica and/or fascioliasis gigantica were just as encouraging as that obtained by Olsen against the former scourge alone using hexachlorethane-bentonite suspension.

The treatment with hexachlorethane (carbon trichloride) and kamala extract occasioned a temporary reduction of milk for a few days; the extract caused a slight to profuse diarrhea which lasted from 2 to 6 days.

The counts per gram of stool in the fifty-two animals ranged from 22 to 244 eggs. Seventeen of them had over 100 ova, the minimum egg-per-gram level set by Taylor (1939) as dangerous for bovine fascioliasis. Owing to the intensity of their infections, six heavily infected cases were given bigger amounts of the flukecide (Table 2) in an attempt to effect a "knock-out" dose without, at the same time, impairing their health. Of the four ruminants that were given the total dose once, two died with all the adult flukes undergoing disintegration; the remainder had livers as clean as a noninfected organ on slaughter. The two emaciated animals given the total amount of the expedient daily for two consecutive days died

[2] Alicata probably dealt with fascioliasis gigantica which is the infection in Hawaii.

together with their parasites three to five days after treatment. The worms were found disintegrated on autopsy.

The expedient seems to be effective also against the conical flukes (*Cotylophoron cotylophorum, Paramphistomum cervi,* etc.) whose eggs were drastically reduced after the medication. The stomachs of the ruminants, however, were not examined, hence the writer could not ascertain whether or not these amphistomes were only sterilized. The effect of hexachlorethane and kamala extract against them deserves further scrutiny.

SUMMARY

The results of treatment with hexachlorethane and kamala extract against fascioliasis hepatica and/or fascioliasis gigantica in fifty-two animals are given in this paper.

In dosis of 10 grams hexachlorethane and 1.75 grams kamala extract per 30 kilos body weight equally distributed over a two-day period, encouraging results were obtained (91.22 per cent efficiency), and the animals generally tolerated the drug well, but not so when the total dose was given only once or when given daily for two consecutive days.

The anthelmintic efficiency of the expedient seems to depend on the intensity of infection. The egg-per-gram level which revealed the absence of worms at autopsy was 44 ova.

Young flukes seem not to be affected by the treatment.

Hexachlorethane-kamala extract combination seems to be a promising remedy also against the conical flukes (*C. cotylophorum, P. cervi,* and others). The effect of this drug against these amphistomes deserves further study.

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SOME FACTORS AFFECTING THE PRODUCTION OF DEXTRAN FROM CANE SUGAR BY LEUCONOSTOC DEXTRANICUM [1]

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TWO PLATES

The production of dextran gum from sucrose (cane sugar) by means of certain organisms has been accomplished by various investigators. The best yield so far recorded is 25 per cent. It required about 2 weeks to produce this amount which is considerably below the theoretical yield of 47.37 per cent.

Recently we had occasion to make some of this gum and incidentally studied the experimental conditions for preparing it. We were successful in working out a method that required only 2 days to produce a theoretical yield. Our results are recorded in this report.

When sucrose ($C_{12}H_{22}0_{11}$) is hydrolyzed it is converted into the two simpler sugars—dextrose ($C_6H_{12}0_6$) and levulose ($C_6H_{12}0_6$). Dextran is a sugar anhydride gum [2] that yields dextrose sugar on hydrolysis. Fernbach, Schoen and Hagiwara,[3] working with *Leuconostoc dextranicum* de Beijerinck, made dextran from sucrose. They found that the organism produced gum only from sucrose, and not from sucrose which was previously hydrolyzed into simpler sugars by acids or invertase, and also not from the isolated dextrose or levulose. Based on the amount of sucrose employed the yield obtained was only about 10 per cent.

[1] This paper was ready for publication September, 1941.

[2] Thaysen, A. C., and L. D. Galloway. The Microbiology of Starch and Sugars (1930) 183.

[3] Comptes Rendus de la Societe de la Biologie 92 (1925) 1418.

Levulosan is also a sugar anhydride gum similar to dextran. It yields levulose sugar on hydrolysis. In 1912 Fernbach and Schoen[4] produced a theoretical yield of levulosan from sucrose by means of bacteria. They showed that the bacteria were able to produce the gum only from nascent levulose that is liberated by the organisms in the hydrolysis of sucrose. The production of levulosan from the levulose part of the sucrose molecule naturally suggested the preparation of dextran from the dextrose portion of the sucrose molecule.

Carruthers and Cooper[5] studied extensively the nutrient requirements and accessory growth factors necessary for a large-scale production of dextran by *Leuconostoc dextranicum* (*Betacoccus arabinosaceous haemolyticus* Kluyver). They found that only a very small amount of gum can be synthesized from glucose alone. The failure to produce dextran from glucose could not have been due to the inhibitory effect of acid produced in the reaction, for the pH of the glucose and sucrose cultures after a week's incubation was practically the same (about 4). After incubating the organisms for 2 weeks at 30° C. with the medium which they developed, these workers were able to synthesize about 25 per cent of dextran based on the sucrose employed. The largest quantity of medium they used for a large-scale production of dextran was 5 liters which was divided into 800-cc portions.

Stacey and Youd[6] followed the method of Carruthers and Cooper for a large-scale production of dextran gum and used the same strain of *Leuconostoc*. They observed unforeseen and inexplicable irregularities in the activity of the organisms. There were growth and also increased viscosity in some flasks, while in others which were prepared in the same manner there was very little or no gum formation. The irregularity became particularly marked when the volume of the culture medium was increased beyond 100 cc and after repeated subculturing of the organisms.

In conformity with the findings of Carruthers and Cooper, Stacey and Youd observed that the acid produced did not have any inhibitory effect on the formation of dextran inasmuch as the pH values of the medium were identical in both viscous and weak cultures during and after growth. Sterilization of sucrose and peptone solutions separately, followed by aseptic

[4] Comptes Rendus Hebdomadaires des Seances de l' Academie des Sciences 155 (1912) 84.

[5] Biochem. Jour. 30 (1936) 1001.

[6] Biochem. Jour. 32 (1938) 1943.

mixing before inoculation, gave increased yields of dextran, but the growth was still irregular.

Stacey and Youd developed a medium for a large-scale production of dextran by using commercial maple syrup for accessory growth substance and for increasing the concentration of sucrose to 20 per cent. The mixed medium was divided into 100-cc portions contained in 500-cc flasks. After they were inoculated with organisms (48 hours old) the cultures were incubated for 10 days at 30° C. The yield of crude gum was 25 per cent based on the sucrose employed.

EXPERIMENTAL PROCEDURE

The *Leuconostoc dextranicum* (*Betacoccus arabinosaceous haemolyticus* Kluyver) which we used in our studies was kindly given to us by Prof. H. J. Kluyver, of Holland. The composition of our culture medium was similar to that developed by Carruthers and Cooper.[7] Our basal medium, designated as medium No. 9 in the experiments, was prepared as follows:

Substitute	Per cent
Sucrose	10.00
Peptone-salt solution:	
Peptone	0.10
Disodium phosphate	0.10
Potassium chloride	0.10
Sodium carbonate	0.013
Distilled water.	
Molasses:	
(50 per cent solution) 5 cc for every 800 cc of the combined liquid medium.	

Double strengths of sucrose (20 per cent) and peptone-salt (0.20 per cent) solutions were sterilized separately in suitable containers. Equal volumes of the cooled solutions were mixed aseptically thus giving a 10 per cent sucrose and 0.10 per cent peptone-salt concentration. To every 800 cc of this mixture 5 cc of molasses (50 per cent) were added.

Preparation of dextran.—The general method for preparing dextran was as follows: Portions (15 cc) of the sucrose-peptone-salt solution containing molasses were poured into sterile calibrated test tubes. The pH of this medium was found by determination to be 7.30–7.70 which was most suitable for the bacteria. Each tube was inoculated with a loopful of

[7] Biochem. Jour. **30** (1936) 1001.

the organisms. After incubation for a definite time the amount of dextran synthesized was determined by adding 3 volumes of alcohol to 1 volume of culture in tarred containers. The gum separated as a whole mass and very little precipitated as powder. The alcoholic mixture was set aside overnight; the supernatant liquid was decanted and the gum was dried in an oven at 100° C. The weight was taken as that of the crude dextran.

The theoretical yield of dextran which can be made from the glucose part of the sucrose molecule may be calculated from the following equations:

$$\underset{\text{sucrose}}{C_{12}H_{22}O_{11}} + H_2O \rightarrow \underset{\text{glucose}}{C_6H_{12}O_6} + \underset{\text{levulose}}{C_6H_{12}O_6}$$

$$\underset{\text{glucose}}{C_6H_{12}O_6} \rightarrow \underset{\text{dextran gum}}{(C_6H_{10}O_5)\,N} + H_2O$$

The molecular weight of sucrose is 342 and that of dextran, 162. Sucrose (342 grams) should yield 162 grams of dextran gum, or a calculated yield of 47.37 per cent.

Effect of water used.—In the first series of experiments medium No. 9 (with distilled water) was used. The tubes were inoculated with organisms (2 days old) and incubated at 30° C. The yield of dextran was low and the growth of the *Leuconostoc* was irregular. Tap water was then used as solvent instead of distilled water in medium No. 9 and the solution was labelled medium No. 10 in the experiments. For comparison two sets of test tubes containing media Nos. 9 and 10, prepared at the same time, were inoculated with the organisms and incubated at 30° C. The amount of dextran synthesized in each medium was determined at various intervals. Results are recorded in Table 1.

TABLE 1.—*Effect of using tap water instead of distilled water in the medium.*

Medium		Gum and pH determined after incubation at 30°C.							
		1 day		2 days		3 days		14 days	
Number	pH.	Gum.	pH.	Gum.	pH.	Gum.	pH.	Gum.	pH.
		Per cent		*Per cent*		*Per cent*		*Per cent*	
9	7.7	4.9	6.50	10.9	8.95	11.1	8.70	11.2	8.20
10[a]	7.65	3.5	6.90	31.4	4.30	32.1	4.30	36.9	3.90

[a] The composition and preparation of medium No. 10 were the same as those of No. 9 except that tap water was used instead of distilled water.

The figures (Table 1) show that the yield of dextran from tap water was higher than that from distilled water, but the theoretical yield was not obtained. The difference in the yields of gum could not have been due to the initial pH of the media as it was practically the same in both cases. The results of the experiments, which will be discussed later, show that the difference was due to certain minerals present in tap water.

Influence of temperature.—To ascertain some other factors which might make possible the complete polymerization of the glucose part of the sucrose molecule the influence of temperature on the activity of the organisms was studied.

One batch of test tubes containing medium No. 10 (pH 7.35) was inoculated with organisms (2 days old) and divided into 3 sets. One set was incubated at 30° C. for one day to allow the organisms to grow and multiply and then incubated at 10° C. The second set of cultures was incubated at 27° C., and the third at 30° C. The amount of gum produced at different incubation temperatures was determined daily. The results are shown in Table 2.

TABLE 2.—*Influence of incubation temperature on the production of dextran.*

Temperature	Gum and pH determined after incubation							
	1 day		2 days		3 days		8 days	
	Gum.	pH.	Gum.	pH.	Gum.	pH.	Gum.	pH.
°C	*Per cent*		*Per cent*		*Per cent*		*Per cent*	
10	19.8	4.35	30.2	4.30	31.8	4.05	48.2	3.86
27	24.7	4.45	49.4	4.10	49.4	3.90	50.5	3.90
30	19.8	4.35	33.8	3.90	36.1	3.67	36.2	3.46

NOTE.—Medium No. 10 (pH 7.35) was used. The culture incubated at 10°C. was first incubated at 30°C. for one day and then transferred at 10°C.

The results (Table 2) show that the theoretical yield of dextran was obtained after a period of 2 days when the organisms were incubated at 27° C. After 8 days, however, the yield of gum from the culture incubated at 10° C. was as high as that kept at 27° C. Both cultures were highly viscous and difficult to pour but the tube kept at 27° C. was more opaque than that incubated at 10° C. The tube kept at 30° C., which was whitish yellow and not very viscous, gave only 36.2 per cent of gum after 8 days of incubation period. These data show that 27° C. is a very suitable temperature for the synthesis of dextran by *Leuconostoc dextranicum*. Longer periods of incubation did

not materially increase the yield of dextran. The amount (49.4 per cent) of crude gum obtained after 2 days of incubation at 27° C. was higher than that of the theoretical yield. This was due, perhaps, to some levulose which was enclosed within the mass of gum when precipitated with alcohol and also, possibly, to the residue of liquid left in the container after decantation.

Age of inoculum.—To determine the proper age of the inoculum, organisms from one culture were inoculated daily in medium No. 10 contained in test tubes and incubated at 27° C. The quantity of gum and pH were determined after 2 days of incubation period, as shown in Table 3.

TABLE 3.—*Age of inoculum and production of dextran.*

Age	Gum and pH determined after 2 days incubation at 27°C		Age	Gum and pH determined after 2 days incubation at 27°C.	
	Gum.	pH.		Gum.	pH.
Days	*Per cent*		*Days*	*Per cent*	
1	50.2	4.45	8	36.6	4.60
2	49.6	4.35	9	35.7	4.75
3	50.1	4.40	10[a]	35.0	4.60
4	50.0	4.30	11	34.0	4.65
5	49.7	4.35	12	27.3	4.65
6	50.0	4.35	13	26.1	4.70
7	49.0	4.20	14	14.2	4.80

[a] Medium No. 10 (pH 7.45) was used.

The results (Table 3) show that an inoculum from 1 to 7 days old can produce the theoretical yield of dextran in 2 days. Older inocula require a longer period of incubation. It was observed, however, that organisms 2 days old gave the best results.

Generations of organisms.—When the organisms were kept for months before being transferred to a new medium, they were too weak to synthesize the theoretical yield of dextran even after very long periods of incubation. Subsequent transfers in liquid medium did not activate them, but when they were grown first in solid medium (medium No. 10 plus 2 per cent agar) and then transferred to liquid medium they became very active again. The first culture in liquid medium, ino-

culated with organisms from a solid medium, was designated as "generation." Subsequent inoculations from one liquid medium to another were designated as "generation 2" and so on (Table 4).

TABLE 4.—*Generations of organisms.*

Generation	Gum and pH determined after 2 days incubation at 27°C.		Generation	Gum and pH determined after 2 days incubation at 27°C.	
	Gum.	pH.		Gum.	pH.
	Per cent			*Per cent*	
1	50.6	4.22	15	47.7	4.30
2	49.7	4.44	16	49.6	4.30
3	48.1	4.40	17	48.7	4.20
4	49.3	4.30	18	48.2	4.35
5	48.4	4.35	19	48.3	4.20
6	48.8	4.35	20	49.1	4.35
7	48.9	4.30	21	48.2	4.30
8	49.9	4.43	22	48.4	4.35
9	48.8	4.51	23	49.6	4.35
10	48.3	4.30	24	49.8	4.30
11	48.3	4.30	25	48.1	4.35
12	48.7	4.36	26	49.2	4.30
13	49.0	3.30	27	49.7	4.35
14	48.1	4.25	28	50.0	4.40

NOTE.—The age of the inoculum was 2 days in all cases.

The data in Table 4 show that subsequent transfers of the organisms in liquid medium did not weaken them nor reduce their ability to polymerize glucose provided the age of the inoculum was 2 days.

Composition of tap water.—Tables 1, 2, and 3 show that by using tap water as solvent, incubating the organisms at 27° C., and using an inoculum 2 days old, the maximum (theoretical) amount of dextran can be produced in 2 days. Analysis of the tap water was obtained from the Metropolitan Water District in order to ascertain the mineral matter which served as nutritive substances for the microörganisms. Table 5 gives the composition of the tap water used in the experiments. Since calcium and magnesium are important mineral consti-

tuents for the metabolism of microörganisms it was thought that perhaps they were responsible for the increase in the amount of gum synthesized by the organisms when tap water was used as solvent.

TABLE 5.—*Chemical analysis of tap water in Manila.*[a]

	p. p. m.
Turbidity	0.15
Color	nil
pH	7.3
Total solids	82.0
Silica (SiO_2)	19.0
Iron and aluminum oxides (R_2O_3)	2.0
Iron (Fe)	traces
Aluminum (Al)	1.0
Calcium (Ca)	13.8
Magnesium (Mg)	4.5
Total alkalinity ($CaCO_3$)	41.0
Acidity (CO_2)	1.5
Bicarbonates (HCO_3)	50.0
Total hardness ($CaCO_3$)	53.0
Sulphates (SO_4)	9.2

[a] This analysis was made in the laboratory of the Balara Filters, Metropolitan Water District.

Calcium and magnesium.—To medium No. 9 (made with distilled water) was added calcium lactate, equivalent to the amount of calcium in tap water. This solution was designated as medium No. 16. To another portion of medium No. 9, magnesium sulphate equivalent to the quantity of magnesium in tap water was added and the solution labelled medium No. 17. To a third portion of medium No. 9 the same amounts of calcium lactate as in medium No. 16 and magnesium sulphate as in medium No. 17 were added together and the solution labelled medium No. 18.

For comparison sets of test tubes containing media Nos. 9, 10, 16, 17, 18 were inoculated with organisms, 2 days old, and incubated at 27° C., and the gum and pH were determined daily. The results are recorded in Table 6.

TABLE 6.—*Calcium and magnesium in the production of dextran.*

Medium No.	Initial pH of medium	Gum and pH determined after incubation at 27°C.					
		1 day		2 days		5 days	
		Gum.	pH.	Gum.	pH.	Gum.	pH.
		Per cent		*Per cent*		*Per cent*	
9	7.53	18.7	4.76	31.3	4.35	31.9	4.00
10	7.45	31.5	4.75	49.0	4.30	49.3	4.25
16	7.60	29.6	4.66	43.9	4.15	44.2	3.95
17	7.50	26.0	4.60	35.2	4.20	35.3	3.85
18	7.35	30.9	5.61	48.3	4.30	48.3	4.10

NOTE.—Medium No. 9 was composed of 10 per cent sucrose; 0.10 per cent disodium phosphate, potassium chloride and peptone; and 0.013 per cent of sodium carbonate dissolved in distilled water. To every 800 cc of the medium 5 cc of molasses (50 per cent) was added.

Medium No. 10 was the same as medium No. 9 except that tap water was used instead of distilled water.

Medium No. 16 was medium No. 9 plus 0.0106 per cent calcium lactate.

Medium No. 17 was medium No. 9 plus 0.00456 per cent of magnesium sulphate.

Medium No. 18 was medium No. 9 plus 0.0106 per cent of calcium lactate and 0.00456 per cent of magnesium sulphate.

Table 6 shows that after 2 days the theoretical yield of dextran was obtained from medium No. 10 while only 31.3 per cent was obtained from medium No. 9. Addition of calcium to medium No. 9 (giving medium No. 16) increased the yield to 43.9 per cent. The addition of magnesium alone to medium No. 9 (giving medium 17) raised the yield to 35.2 per cent. When calcium and magnesium were added together to medium No. 9 (giving medium 18) the yield of dextran was increased by 17 per cent. This is about equal to the sum (16.5 per cent) of the increases due to calcium and magnesium (media Nos. 16 and 17) added separately. Calcium and magnesium appear to be essential mineral factors in the synthesis of dextran from sucrose by *Leuconostoc dextranicum.*

Importance of nascent dextrose.—A sample of dextrose crystals prepared by the Insular Sugar Refining Company, Manila, was kindly presented to us by the superintendent, Mr. J. E. Mahoney. This sample was used in 5 and 10 per cent concen-

trations instead of sucrose in some of our media. The tubes containing the media were inoculated with organisms 2 days old, and the cultures were incubated at 27° C. After 2 days there was no gum formation. The cultures were further incubated for a period of one week and there was still no evidence of dextran formation. These results confirm the findings of Fernbach, Schoen, and Hagiwara [8] and also of Carruthers and Cooper [9] that dextran can be synthesized only from nascent glucose which is liberated from sucrose by the organism itself.

Comparative dextran production.—Comparative results obtained by different investigators on the production of dextran are given in Table 7.

TABLE 7.—*Comparative results obtained by different investigators on the production of dextran.*

Investigators	Incubation		Yield of crude dextran[a]
	Temperature	Period	
	°C.	*Days*	*Per cent*
Fernbach, Schoen, and Hagiwara (1925)[b]			10
Carruthers and Cooper (1936)[c]	30	14	25
Stacey and Youd (1938)[c]	30	10	25
Baens-Arcega and Yenko (1941)[c]	27	2	47.5–50.6

[a] The yield of crude dextran was computed on the amount of sucrose employed.
[b] *Leuconostoc destranicus* de Beijerinck was used.
[c] *Leuconostoc dextranicum* (*Betacoccus arabinosaceous haemolyticus* Kluyver) was used.

The data given in Table 7 show that Fernbach, Schoen, and Hagiwara obtained 10 per cent of dextran based on the sucrose employed. Carruthers and Cooper, as well as Stacey and Youd, succeeded in increasing the yield to 25 per cent after incubating the organisms for about 2 weeks. In our investigations we produced in 2 days 47.5–50.6 per cent of dextran, which is about the theoretical yield, by incubating the organisms at 27° C., and using our medium. The same yield was obtained when we worked with a fairly large volume of medium (50 liters at one time) distributed in 4-liter Erlenmeyer flasks.

Appearance of the organisms.—Smears of the organisms were stained in the following manner:

A loopful of diluted culture was placed on a clean slide, smeared, and fixed by drying over a small flame. It was

[8] Comptes Rendus de la Societe de la Biologie 92 (1925) 1418.
[9] Biochew. Jour. 30 (1936) 1001.

stained with carbol fuchsin solution for 2 to 5 minutes with the aid of heat. The stained organisms were rinsed with distilled water and dried over a flame. A loopful of saturated nigrosine NB solution was placed on one end of the slide and spread over the smear with the aid of the edge of another slide. Rapid drying was necessary to avoid decolorizing the organisms.

Under the high-power objective of the microscope the organisms appeared red surrounded by huge white capsules against a bluish background. They appeared singly, sometimes in diplos (pairs) and occasionally in short chains. The capsules of the organisms grown in solid medium were larger (Plate 1, fig. 2) than those grown in liquid medium (Plate 1, fig. 1).

When seen under the oil-immersion lens (Plate 2, figs. 1 and 2) two or more organisms were often found enclosed within the capsule. Capsules of organisms grown in solid medium contained more cells (Plate 2, fig. 2) than those grown in liquid medium (Plate 2, fig. 1). This fact recalls the observation of Mendes, as cited by Taar and Hibbert,[10] that inside the gelatinous capsules of *Leuconostoc mesenteroides* small cells were able to multiply by fission. This observation contributes additional and more conclusive evidence supporting the assumption that the mucilaginous fermentation results from the activity of the microörganisms.

Since the individual organisms enclosed within the capsules were clearly defined only under the oil-immersion lens, measurements of the organisms grown in liquid medium were made under this magnification. The cells within the capsules had an average of 0.9 micron in diameter. The size of the capsules varied with the number of organisms enclosed. Measurements of capsules enclosing single cells were taken. These capsules had an average size of 2.6 microns in width and 3.5 microns in length.

The gum was purified from the thick medium by precipitating it with alcohol. The white mass was dissolved in water, precipitated with alcohol a second time, and dried in a vacuum oven. A small portion of the purified gum was dissolved in water and smears were stained. The same capsulated organisms were seen.

[10] Canad. Jour. of Res. 5 (1931) 419.

According to Jrgensen, Hansen, and Lund,[11] the slime capsule formed by *Betacocci* consists of a monosaccharide anhydride called dextran.

Bergey,[12] in describing the species of *Leuconostoc mesenteroides* (Cieukowski) Van Tieghem, states that the chains of these organisms are surrounded by a thick, gelatinous, colorless membrane consisting of dextran.

The capsules of *Leuconostoc dextranicum* may likewise be composed of dextran.

Capsule formation and temperature.—In our low-temperature experiments (Table 2) the organisms were first incubated at 30° C. for one day to allow them to grow and multiply. Very little change was noted in the inoculated medium which was not viscous and only slightly cloudy. The culture was then transferred to 10° C. After one day at this temperature it became very viscous and transparent. The viscosity would naturally suggest the formation of considerable gum; however, when precipitated with alcohol, the yield of dextran was only 30.2 per cent as the material was partly soluble in alcohol.

The low temperature might have stimulated the organisms to form a protective coating or capsule. This coating may have consisted of dextran together with a soluble constituent (an intermediate product in the synthesis of dextran). Attempts to observe the organisms at this stage were not successful as it was difficult to stain the capsules.

The synthesis of dextran proceeded slowly and after 8 days at 10° C. the yield gradually increased to 48.2 per cent, which is about the theoretical amount.

A very suitable temperature for these organisms is apparently 27° C. When they were incubated at this temperature for 2 days 49.4 per cent of dextran was obtained. Under these conditions the organisms were not exposed to an unfavorable low temperature which might cause a retarding action. The culture was opaque and not thick as in the low-temperature experiment. The main activity at the optimum temperature is the synthesis of dextran.

[11] Jrgensen, A., A. Hansen, and A. Lund. Microörganisms and Fermentation (1939) 336.

[12] Bergey, David H. Bergey's Manual of Determinative Bacteriology (1930) 64.

When the organisms were incubated at 30° C., the temperature was too high for the proper activity of the organisms since the amount of dextran synthesized was not as much as that formed at lower temperatures.

SUMMARY

Dextran is a gum synthesized from the glucose part of the sucrose molecule by *Leuconostoc dextranicum* (*Betacoccus arabinosaceous haemolyticus* Kluyver).

The experimental conditions for the preparation of dextran from sucrose were investigated.

A suitable medium for the microörganisms to produce the theoretical yield (47.37 per cent) was developed. This medium consisted essentially of solutions of sucrose, peptone, alkali and alkali earth salts with a trace of molasses.

The optimum temperature for the production of dextran was found to be 27° C.

Experiments showed that an inoculum 1 to 7 days old can produce the theoretical yield of dextran in 2 days when the organisms were incubated at the optimum temperature.

Weakened organisms may be activated by growing them in a solid medium and then transferring them to a liquid medium.

Subsequent transfers of the microörganisms in liquid medium did not affect their activity provided the age of the inoculum was 2 days.

Tap water gave better results for preparing the medium than distilled water. The calcium and magnesium in tap water were found to be necessary nutrient factors for *Leuconostoc* in the synthesis of dextran.

Our experiments showed that dextran can be synthesized only from nascent glucose which is liberated from sucrose by the organism itself. When dextrose was used instead of sucrose, as carbohydrate material in the medium, dextran was not produced.

Reference was made to the comparative results obtained by different investigators on the production of dextran.

Carruthers and Cooper were able to produce 25 per cent of dextran based on the amount of sucrose employed by incubating the microörganisms for 2 weeks.

By using our medium we succeeded in synthesizing the theoretical yield of dextran (47.37 per cent) in 2 days. The

largest volume of medium we employed at one time was 50 liters, distributed in 4-liter Erlenmeyer flasks.

Photomicrographs of the stained capsules of *Leuconostoc*, grown in liquid and solid media, as observed under the high-power and also the oil-immersion objectives, were made. The capsules contained one or more cells as observed under the oil-immersion lens. Those enclosing single cells of organisms grown in liquid medium had an average size of 2.6 microns in width and 3.5 microns in length.

Our investigation indicates that the capsule is probably composed of dextran.

ILLUSTRATIONS

Plate 1

Fig. 1. *Leuconostoc dextranicum* grown in liquid medium as seen under the high-power objective; × 700.

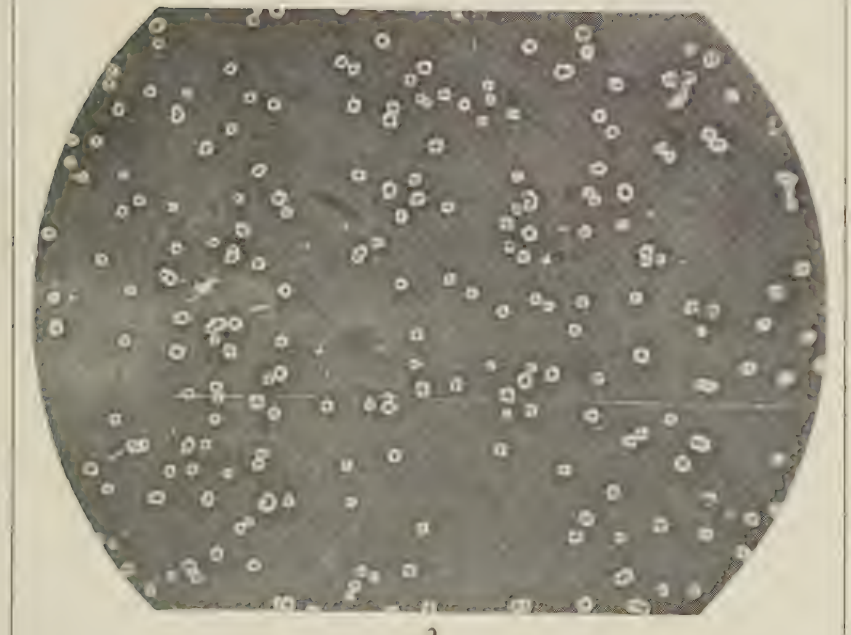
2. *Leuconostoc dextranicum* grown in solid medium as seen under the high-power objective; × 625.

Plate 2

Fig. 1. *Leuconostoc dextranicum* grown in liquid medium as seen under the oil-immersion lens; × 1,510.

2. *Leuconostoc dextranicum* grown in solid medium as seen under the oil-immersion lens; × 1,100.

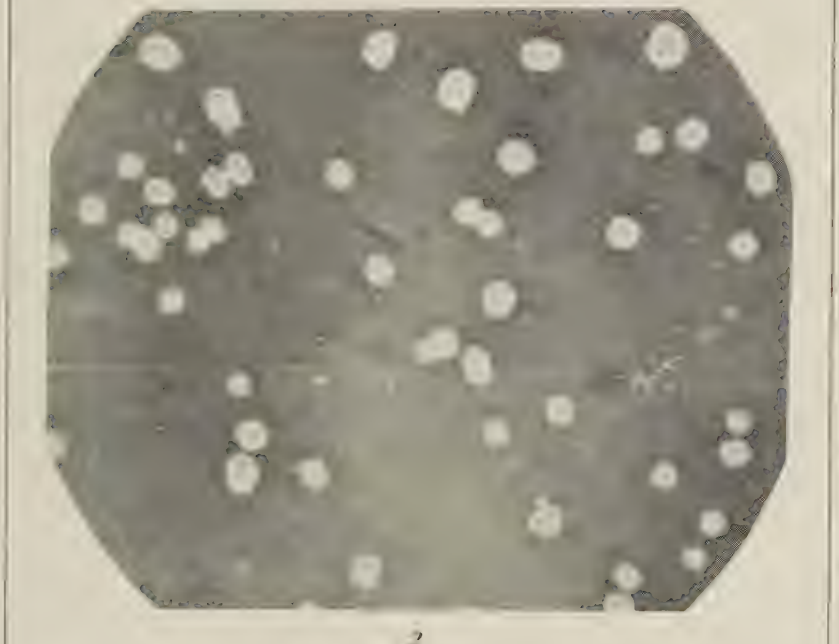
1



2

PLATE 1.

1



2

PLATE 2.

JATROPHA CURCAS LINN. (TUBA) AS A SOURCE OF NATURAL DYE [1]

By MAGDALENA R. ALDE, FRANCISCO AGCAOILI, and ROSA J.-COCHICO

Of the Bureau of Science, Manila

Jatropha curcas Linn., known as *tuba* in Tagalog, *taua-taua* in Ilocano, *tuba-tuba* in Visayan, is found in thickets and hedges throughout the Philippines.[2] It is common all the year in and about towns, and has been used for various purposes. The natives make use of the oil from the nuts for lighting their houses. It has been found also that almost all parts of this plant could be used for medicinal purposes.[3] It was observed that the decoction from the leaves and branches which were used for curing purposes, left a more or less permanent stain on the cloth. This fact has led the writers to study it as a source of natural dye, and to determine the proper method of applying the dye to ensure evenness and fastness qualities so that our local weavers and dryers can utilize it as a substitute for synthetic dyes.

METHODS OF EXTRACTION

Two methods of extraction, the simplest possible in order to make it easy for local dyers to apply them in their respective localities, were tried in extracting the coloring matter from the leaves and stems of the tuba plant. These methods are as follows:

Procedure 1.—The leaves and tender stems of the tuba were boiled for 4 hours. The solution was filtered through a cheesecloth and later concentrated into a syrupy consistency

[1] This paper was started before the outbreak of the war, but owing to a number of circumstances its completion has been delayed.

[2] Merrill, Elmer D., Flora of Manila (1912) 290.

[3] Brown, William H., Minor Products of Philippine Forests 3 (1921) 200.

by evaporation. The concentrate was a yellowish-olive syrupy substance.

Procedure 2.—The same procedure as in 1 was followed with the exception that the evaporation was continued to dryness. The concentrate was further dried in an oven. The dried extract obtained was in the form of blackish-brown lumps.

The extract obtained from the above procedures, however, included some impurities in it. In the succeeding experiments it was used in the dyeing of cotton. Several ways of applying it to cotton were tried, and the dyed material was tested for its fastness properties.

PROPERTIES OF THE EXTRACT

The dried extract has a blackish-brown appearance and is in the form of lumps. It is soluble in water, and readily soluble in hot water, having a brownish color in solution. When hydrochloric acid and sulfuric acid were added to the extract, its color is slightly changed. With sodium hydroxide the color turns deep brown and the extract is more readily soluble by its presence.

PRELIMINARY TREATMENT OF COTTON

Raw cotton goods contain waxes, serecins, oils, and other impurities. These impurities must be removed before the cotton goods are dyed, if good penetration and level dyeing are to be obtained.

The cotton yarn is scoured or boiled in a bath containing 10 per cent sodium carbonate (2 per cent sodium hydroxide can also be used) on the weight of the material. The material is worked in this bath for 2 hours or left overnight in the above solution after thorough wetting with water. It is then rinsed well with water and hydroextracted.

METHODS OF DYEING

Various methods of applying the natural dyes on cotton were tried. These dyes gave different shades of tan and brown. Both extracts obtained by the two procedures of extraction were used and the dyed material was tested for its fastness properties.

DYEING WITH THE TUBA CONCENTRATE

METHOD 1

The scoured cotton yarn was dyed in a bath containing the tuba concentrate diluted with water enough to cover the yarn. This was brought to the boil and worked for ¾ to 1 hour. Then the dyed yarn was hydroextracted.

Several after-treatments were tried on the dyed material.

After-treatment (*a*).—The dyed yarn was after-treated with a warm solution containing 3 per cent alum for 30 minutes. Then it was rinsed and dried.

After-treatment (*b*).—The dyed yarn was immersed in a solution containing 4 per cent lead subacetate for half an hour and subsequently rinsed and dried.

After-treatment (*c*).—The dyed yarn was immersed in a solution containing 1 per cent copper sulphate and 1 per cent potassium dichromate for half an hour. Then it was rinsed and dried.

After-treatment (*d*).—The dyed yarn was immersed in a warm bath containing 2 per cent ferric chloride for about 30 minutes. Then it was rinsed and dried.

After-treatment (*e*).—The dyed material was immersed in a bath containing 3 per cent sodium sulphide for 30 minutes, and then was rinsed and dried.

After-treatment (*f*).—The dyed material was immersed in a bath containing 3 per cent chromium fluoride for 30 minutes. Then it was rinsed and dried.

METHOD 2

The scoured yarn was dyed in a bath containing the tuba coloring matter [0.4 per cent sodium hydroxide and 10 per cent of common salt (sodium chloride)]. This was worked in the bath for ¾ to 1 hour and brought to the boil. Then it was hydroextracted.

After-treatment.—The dyed material was immersed in a bath containing 3 per cent copper sulphate for 30 minutes. Then it was rinsed and dried.

DYEING WITH THE TUBA DRIED EXTRACT

METHOD 1

The scoured yarn was dyed in a bath containing the tuba dried extract and sufficient water to keep the yarn immersed. This was brought to the boil and worked for ¾ to 1 hour.

It was then hydroextracted, and several after-treatments were applied.

After-treatment (*a*).—The dyed yarn was immersed in a solution containing 3 per cent copper sulphate for 30 minutes. Then it was rinsed and dried.

After-treatment (*b*).—The dyed yarn was immersed in a bath containing 3 per cent copper sulphate, and 3 per cent potassium dichromate for 30 minutes. Then it was soaped, rinsed, and dried.

After-treatment (*c*).—The dyed yarn was immersed in a solution containing 4 per cent lead subacetate for 30 minutes. This was soaped, rinsed, and dried.

After-treatment (*d*).—The dyed yarn was immersed in a solution containing 3 per cent chromium fluoride for 30 minutes. Then it was rinsed and dried.

After-treatment (*e*).—The dyed yarn was immersed in a solution containing 3 per cent potassium dichromate for 30 minutes. Then it was rinsed and dried.

METHOD 2

The second yarn was dyed in a bath containing 30 per cent tuba dried extract, 0.4 per cent sodium hydroxide, 10 per cent common salt (sodium chloride), and sufficient water to keep the yarn immersed. This was brought to the boil and worked in this dye bath for ¾ to 1 hour. Then it was hydroextracted.

The following after-treatments were applied:

After-treatment (*a*).—The dyed yarn was immersed in a bath containing 3 per cent ferric chloride for 30 minutes. Then it was rinsed and dried.

After-treatment (*b*).—The dyed yarn was immersed in a bath containing 3 per cent alum for 30 minutes. Then it was rinsed and dried.

After-treatment (*c*).—The dyed yarn was immersed in a bath containing 3 per cent potassium dichromate for half an hour. Then it was rinsed and dried.

Different shades of tan were obtained from the dried coloring matter and light shades of brown from the concentrate. The

shades, however, depended upon the amount of coloring matter used.

METHOD 3

The scoured yarn was dyed in a bath containing 30 per cent of the dried extract, 3 per cent ferric chloride and sufficient water to cover the yarn. This was brought to the boil gradually and worked for ¾ to 1 hour.

After-treatment.—It was then after-treated in a solution containing 4 per cent potassium dichromate for 30 minutes. Then it was soaped, rinsed, and dried.

FASTNESS PROPERTIES

The dyed materials were tested for their fastness properties. Fair results were obtained from them. Tables 1 and 2 show the fastness properties of these dyed yarns. The fastness is graded according to the following numbers: 1, excellent; 2, very good; 3, good; 4, moderate; 5, poor.

TABLE 1.—*Fastness properties of cotton yarn dyed with tuba concentrate.*

(Procedure 1)

[1, Excellent; 2, very good; 3, good; 4, moderate; 5, poor.]

Methods of dyeing	Light	Rubbing	Washing	Lime water	Soda boil	Perspiration	Alkalies		Acetic acid
							10 per cent Na_2CO_3.	Ammonia	
Method 1:									
(*a*) Alum, 3 per cent	3	1	3	3	3	2	3	2	1
(*b*) Lead subacetate, 4 per cent.	4	1	4	3	4	4	3	3	3
(*c*) Copper sulphate, 1 per cent. Potassium dichromate, 1 per cent.	2	1	3	3	3	3	3	2	3
(*d*) Ferric chloride, 2 per cent.	3	1	3	3	3	4	3	1	4
(*e*) Sodium sulphide, 3 per cent.	3	1	3	3	3	3	3	3	3
(*f*) Chromium fluoride, 3 per cent.	3	1	3	3	3	2	3	1	2
Method 2:									
Copper sulphate, 3 per cent.	3	1	3	3	3	3	3	1	2

TABLE 2.—*Fastness properties of cotton yarn dyed with tuba dried extract.*

(Procedure 2)

[1, Excellent; 2, very good; 3, good; 4, moderate; 5, poor.]

Methods of dyeing	Light	Rubbing	Washing	Lime water	Soda boil	Perspiration	Alkalies		Acetic acid
							10 per cent Na_2CO_3.	Ammonia	
Method 1:									
(*a*) Copper sulphate, 3 per cent.	4	1	4	3	3	3	3	3	2
(*b*) Potassium dichromate, 3 per cent. Copper sulphate, 3 per cent.	3	1	3	3	3	3	3	1	2
(*c*) Lead subacetate, 4 per cent.	5	2	3	3	3	3	2	3	3
(*d*) Chromium fluoride, 3 per cent.	4	1	3	3	3	3	3	2	2
(*e*) Potassium dichromate, 3 per cent.	5	1	3	3	3	3	3	2	2
Method 2:									
(*a*) Ferric chloride, 3 per cent.	5	1	4	4	4	4	3	4	4
(*b*) Alum, 3 per cent	5	1	3	2	3	3	3	1	2
(*c*) Potassium dichromate, 3 per cent.	5	1	4	4	4	4	3	2	3

SUMMARY

1. The coloring matter of the leaves and stems of *Jatropha curcas* Linn. (tuba) was extracted by boiling with water, one extract evaporated to a syrupy consistency, and the other, to dryness.

2. The extracted matter was applied to cotton yarn by different methods of dyeing and after-treatment.

3. The dyed cotton yarn was tested for its fastness properties.

4. Fair results were obtained from these experiments.

NOTES ON THE INSECT FAUNA OF THE SAMAR GROUP, PHILIPPINES

By F. F. Bibby
Of Smithville, Mississippi

The material on which the list is based was collected off hours while the writer was stationed as a member of a U. S. Navy malaria and epidemic control unit on Calicoan Island from April to October, 1945.

Besides the writer, J. R. Dodds, L. E. Fronk, J. L. Imhof, Henry Staller, and J. W. Stinson, all of the malaria and epidemic control unit, contributed material and assisted otherwise. Other Navy personnel who contributed material were: H. J. Rayner, J. G. Spann, A. W. Rowbottom, R. C. Hartsfield, and a Mr. Ties.

The identification of the insects, except the Asilidæ, was made by the United States Department of Agriculture, Agricultural Research Administration, Bureau of Entomology and Plant Quarantine, Washington, D. C. The identification of the Asilidæ was made by the Bartlett Tree Research Laboratories, Stamford, Connecticut.

The identification of the plants included in the list was made by the United States Department of Agriculture, Agricultural Research Administration, Bureau of Plant Industry, Soils, and Agricultural Engineering, Beltsville, Maryland.

The specimens were taken on Calicoan Island and at nearby places on the adjacent islands of Samar and Leleboon, all between the Pacific Ocean and Leyte Gulf. The elevation varied from seal level to 750 feet above, with some rather abrupt changes.

Some notes on the flora follow:

Wild mallows: *Urena lobata, Sida rhombifolia, Hibiscus tiliaceus, Thespesia populnea, Abutilon* sp.

Other wild plants: *Morus* sp., *Callicarpa* sp., ebony, mahogany, acacia, poinsettia, *Passiflora* sp., cycads, ferns, pandanus, verbena, bamboo, fishtail palm, *Anamirta cocculus* (*lagtang* or fish berry), *Barringtonia asiatica* (fish poison), *Amaranthus* sp., *Polanisia icosandra,* morning-glory (Convolvulaceæ), *Ficus* spp.

Food plants: breadfruit, banana, guava, citrus, coconut, cassava, papaya, taro, sweet potato.

Ornamentals: *Hibiscus rosa-sinensis, Malvaviscus arboreus, Codiaeum variegatum, Abelmoschus moschatus, Bougainvillea, Delonix regia, Datura alba, Lochnera rosea.*

Other cultivated plants: *Derris* sp., cotton (occasional stalk for wicks), tobacco.

In the list of insects to follow, there are represented 13 orders, 100 families, 246 genera, and 310 species.

The number of species to an order, to a family, and to a genus, or the absence of any group, is not necessarily indicative of relative abundance. It could have been affected by facility to collect, by facility to send for determination, or by preference of the collectors.

However, scarcity of species accounts for the absence of the followning groups from the list:

Carabidæ
Meloidæ
Mutillidæ
Thysanoptera.

ANOPLURA

HAEMATOPINIDÆ

Hoplopleura sp.—Calicoan Island, July 2, 1945, from rat.

COLEOPTERA

ANOBIIDÆ

Lasioderma sp., prob. *serricorne* Fabricius—Calicoan Island, May, 1945.

ANTHRIBIDÆ

Undet. sp.—Calicoan Island, August 27, 1945, from blooms of *Hibiscus tiliaceus.*

BOSTRICHIDÆ

Dinoderus minutus (Fabricius)—Guiuan, Samar, from wooden-soled sandals.

Xylopsocus capucinus (Fabricius)—Calicoan Island, August 29, 1945.

Xylothrips flavipes (Ill.)—Calicoan Island, October 8, 1945, from man who reported it had bitten him.

BUPRESTIDÆ

Agrilus occipitalis Eschscholtz—Calicoan Island, October 15, 1945, from grapefruit foliage.

Chrysodema smaragdula Olivier—Calicoan Island, spring of 1945.

Sambus sp.—Calicoan Island, October 10, 1945, from foliage of shrub along Leyte Gulf. Numerous and lively.

CANTHARIDÆ

Tylocerus atricornis (Guér.)—Calicoan Island, May and June, 1945, from vegetation.

CERAMBYCIDÆ

Aeolesthes induta Newmann—Calicoan Island, spring of 1945.

Apomecyna quadrifasciata Thomson—Calicoan Island, spring and summer of 1945, at light.

Batocera rubus var. *miniszechi* Thomson—Calicoan Island, spring and summer of 1945; one collected August 12 bore *Lophochernes* sp., possibly new (Arachnida, Cheliferidæ).

Cacia vermiculata ab. *dissoluta* Heller—Calicoan Island, June 27, 1945, from jungle vegetation about 500 feet above sea level.

Ceresium sp.—Calicoan Island, September 1, 1945, indoors.

Daphisia leopoldi Fisher—Calicoan Island, spring of 1945.

Dihammus pseudobianor Breun. ?—Calicoan Island, August 27, 1945, from jungle.

Glenea gracilis Aurivillius—Calicoan Island, August 10, 1945, from jungle.

G. maura Pascoe—Calicoan Island, spring of 1945.

G. suavis Newmann—Calicoan Island, May, 1945.

G. versuta ab. *fasciolata* Aurivillius—Calicoan Island, August 10, 1945, from jungle.

G. sp.—Calicoan Island, spring of 1945.

Ichthyodes biguttula Newmann—Calicoan Island, May, 1945.

Lachnopterus auripennis (Newmann)—Calicoan Island, May to July, 1945.

Nyctimene ochraceovittata Aurivillius—Calicoan Island, May, 1945.

Ostedes pauperata Pascoe—Calicoan Island, May, 1945, from jungle.

Pothyne trivittata Newmann—Calicoan Island, June, 1945.

CHRYSOMELIDÆ

Acrocrypta cumingi (Baly)—Calicoan Island, October 18, 1945, from vegetation, 300 feet above sea level.

Aulacophora sp., perhaps a variety of *A. rosae* (Fabricius)—Calicoan Island, May and June, 1945, common on jungle vegetation.

Colasposoma sp., prob. *cumingi* Baly—Calicoan Island, August 5, 1945, from jungle.

C. gregarium LeF.—Calicoan Island, May, 1945.

C. sp.—Calicoan Island, August 5, 1945, from jungle.

Dactylispa sp., new to collection at Washington—Calicoan Island, May, 1945.

Laccoptera luzonica Spaeth—Guiuan, Samar, April, 1945, from *Abelmoschus moschatus.*

Metriona disphorica Spaeth—Calicoan Island, May, 1945, from jungle.

M. trivittata (Fabricius)—Calicoan Island, May, 1945.

Nodosocantha sp., prob. *sexnotata* (Weise)—Calicoan Island, August 10, 1945, from jungle.

Phytorus, 2 spp.—Calicoan Island, May and August, 1945, from jungle.

Platypria sp., new to collection at Washington—Calicoan Island, May, 1945.

Rhyparida sp.—Samar, April, 1945.

Sermyloides sp.—Calicoan Island, May 7, 1945.

Xenoda sp. near *pallida* Jac.—Calicoan Island, April, 1945.

Undet. sp. of genus near *Aulacophora*—Calicoan Island, July 26, 1945, common.

Undet. sp., perhaps *Phytorus* sp., new to collection at Washington—Calicoan Island, October 10, 1945, from foliage of *Thespesia populnea* along Leyte Gulf.

Undet. sp. of genus near *Sphaeroderma*—Calicoan Island, August 10, 1945, from jungle.

Undet. sp. of Galerucinæ, new to collection at Washington—Calicoan Island, August 13, 1945, feeding on foliage of a baylike tree near Leyte Gulf.

CICINDELIDÆ

Cicindela lacrymosa Dej.—Calicoan Island, May, 1945, from sand in the open.

Collyris sp.—Calicoan Island, May, 1945, from jungle vegetation.

Neocollyris sp.—Calicoan Island, August 13, 1945, from jungle vegetation.

Therates labiatus fulvipennis Chd.—Calicoan Island, May to October, 1945, from jungle vegetation; alert but easily captured.

Tricondyla conicicollis Chd.—Calicoan Island, May to July, 1945, from jungle vegetation.

T. punctipennis Chev.—Calicoan Island, May, 1945, from jungle vegetation.

T. sp.—Calicoan Island, May, 1945, from jungle vegetation.

COCCINELLIDÆ

Catana sp., perhaps *clauseni* Chapin—Calicoan Island, October 15, 1945, predator of *Tenaphalara fascipennis* (Crawford) (Psyllidæ) on rubberlike shrub, 250 feet above sea level.

Coelophora 8-punctata (Fabricius)—Calicoan Island, June, 1945, predator of *Aphis medicaginis* Koch on a forage legume (sonting).

C. sp.—Calicoan Island, May, 1945, from jungle vegetation.

Epilachna n. sp.—Calicoan Island, June, 1945, from jungle vegetation.

E. sp.—Calicoan Island, May 7 and August 10, 1945, from jungle vegetation.

Serangium sp.—Calicoan Island, October 15, 1945, in association with *Catana* sp. preying upon *Tenaphalara fascipennis* (Crawford) (Psyllidæ) on a rubberlike shrub (not *Ficus*) 250 feet above sea level.

Undet. sp. of *Scymnus* or related genus—Calicoan Island, August 5, 1945, from underside of leaf of jungle plant of the taro (elephant's-ear) group.

CUCUJIDÆ

Ahasverus advena (Waltl.)—Calicoan Island, June, 1945, at light.

Silvanus bidentatus (Fabricius)—Calicoan Island, June, 1945, numerous and a nuisance, at light.

CURCULIONIDÆ

Alcidodes sp.—Guiuan, Samar, from foliage, October 17, 1945.

Amorphoidea sp., probably same as species treated as *lata* Mots. by Otanes and Butac (1939)—Calicoan Island and Samar, May to October, 1945, larvæ in seed pods of *Hibiscus tiliaceus* and *Thespesia populnea*, and adults numerous in blooms of both hosts.

Apion sp.—Calicoan Island, May, 1945, from foliage of *Urena lobata.*

Homalocyrtus sp.—Calicoan Island, May 10, 1945, from foliage of *Hibiscus tiliaceus.*

Metapocyrtus sp.—Calicoan Island, October 18, 1945, on foliage 200 feet above sea level.

Pachyrhynchus sp.—Samar, May, 1945.

Peribleptus dealbatus (Boisduval)—Calicoan Island, June, 1945, from jungle vegetation.

Pyrgops sp.—Samar, September 8, 1945, from foliage of *Urena lobata,* and Calicoan Island, September 26, from foliage of *Hibiscus tiliaceus.*

Rhynchites plagiocephalus Voss—Calicoan Island, October 15, 1945, from foliage.

Rhynchophorus ferrugineus (Olivier) or *pascha* Boh.—Calicoan Island, August 20, 1945.

Undet. sp. of Celeuthetini—Calicoan Island, May 7, 1945, from foliage of *Hibiscus tiliaceus* and from foliage of pepper.

DYTISCIDÆ

Hydaticus fabricii (McLeay)—Calicoan Island, May 14, 1945, from standing water in swamp.

ELATERIDÆ

Agrypnus bifoveatus Candèze—Calicoan Island, June, 1945, at light.

Neodiploconus sp.—Calicoan Island, May 7, 1945.

EROTYLIDÆ

Hybosoma hydropicum Gorh.—Calicoan Island, June 27, 1945, from jungle, 500 feet above sea level.

Rhopalotritoma amabilis Heller—Calicoan Island, from jungle, 300 feet above sea level.

LAMPYRIDÆ

Luciola sp.—Calicoan Island, May 8, 1945.

LANGURIIDÆ

Anadastus sp.—Calicoan Island, June, 1945.

LYCIDÆ

Lyropaeus sp.—Calicoan Island, October 18, 1945, from vegetation, 300 feet above sea level.

Metriorhynchus sp.—Calicoan Island, May, 1945.

Undet. sp., genus not recognized—Calicoan Island, August 19, 1945, from jungle vegetation.

MORDELLIDÆ

Glipa sp.—Calicoan Island, May and June, 1945, on jungle vegetation, common but evasive.

NITIDULIDÆ

Carpophilus dimidiatus (Fabricius)—Calicoan Island, July 2, 1945, combed from rat trapped in commissary.

Haptoncus sp. near *luteolus* Er.—Calicoan Island, September 9, 1945, from blooms, flower buds, and seed pods of *Hibiscus tiliaceus;* and Samar, September 10, from same kind of material.

Undet. sp., not in U. S. National Museum—Calicoan Island, September 6, 1945, from fresh and wilted blooms of *Thespesia populnea;* and Samar, September 8, from blooms of *Hibiscus tiliaceus.*

PLATYPODIDÆ

Platypus sp., near *lepidus* Chap.—Calicoan Island, August 29, 1945, indoors.

SCARABAEIDÆ

Anomala (*Euchlora*) *chloropyga* Burmeister—Calicoan Island, May and June, 1945, from jungle vegetation, 300 feet above sea level.

A. sp.—Calicoan Island, October 15, 1945, from jungle vegetation.

Dasyvalgus panaonus Mos.—Calicoan Island, October 18, 1945, from jungle vegetation, 300 feet above sea level.

Microserica sp.—Samar, October 17, 1945, from foliage.

Onthophagus sp.—Calicoan Island, June, 1945.

Oryctes rhinoceros (Linnæus)—Samar, May, 1945.

Philaelota sulana Heller—Calicoan Island, August 15, 1945, indoors.

Pseudomalaia semperi Kraatz—Guiuan, Samar, April, 1945, from blooms of *Abelmoschus moschatus.*

SCOLYTIDÆ

Xyleborus sp., prob. *parvulus* Eichhoff—Calicoan Island, June, 1945, indoors at light.

X. sp., prob. *perforans* (Woll.)—Calicoan Island, June, 1945, indoors at light; and July 4, 1945, combed from a trapped rat.

X. sp.—Calicoan Island, May, 1945, reported to have bitten a person.

TENEBRIONIDÆ

Ceropria sp.—Calicoan Island, October 10, 1945, indoors.

Strongylium sp.—Calicoan Island, October 18, 1945, from vegetation, 300 feet above sea level.

Undet. sp. of *Bradymerus* or related genus—Calicoan Island, August 7, 1945, from water in axil of banana leaf.

COLLEMBOLA

ISOTOMIDÆ

Isotomurus sp.—Calicoan Island, July, 1945, from water puddle accumulated from recent rain.

CORRODENTIA

PSOCIDÆ

Ectopsocus sp.—Calicoan Island, July, 1945, from pasteboard box containing dry buds of *Hibiscus tiliaceus.*

TROGIIDÆ

Liposcelis sp.—Calicoan Island, June, 1945, devouring museum specimens of mosquitoes.

DIPTERA

AGROMYZIDÆ

Desmometopa sp.—Calicoan Island, July 25, 1945, in association with *Hecamede* sp., prob. *persimilis* Hendel, and *Gymnopa* sp. (Ephydridæ).

Milichiella sp.—Calicoan Island, September 7, 1945, from tip of twigs of *Thespesia populnea.*

Tethina sp.—Calicoan Island, July 23, 1945, in association with *Hebecnema* sp. (Ephydridæ) on seaweed along shore of the Pacific Ocean; and September 12, indoors.

ASILIDÆ

Dalmalina semperi O. S.—Calicoan Island, August 10, 1945, from fermenting banana plant.

D. sp.—Calicoan Island, May and June, 1945.

Maira sp.—Calicoan Island, June and August, 1945.

Ommatius chinensis Fabricius—Calicoan Island, June 7, 1945.

O. sp.—Calicoan Island, June 7, 1945.

Philodicus longipes Schiner—Calicoan Island, June, 1945, one with prey, small butterfly (Lycaenidæ); and Leleboon Island, June 22, 1945.

Promachus bifasciatus Macquart—Leleboon Island, June 22, 1945.

P. manilliensis Macquart—Calicoan Island, May, 1945.

P. philippinus Ricardo—Calicoan Island, May, 1945.

P. varipes Macquart—Calicoan Island, May, 1945.

P. sp.—Calicoan Island, August 11, 1945.

BOMBYLIIDÆ

Undet. sp., prob. of genus *Hyperalonia*—Leleboon Island, June 26, 1945.

CALLIPHORIDÆ

Chrysomya megacephala (Fabricius)—Calicoan Island, May, 1945; and Leleboon Island, June 25, 1945.

Hemipyrellia tagaliana (Bigot)—Calicoan Island, May 15, 1945.

CHLOROPIDÆ

Eutropha n. sp., near *noctilus* (Walker)—Calicoan Island, July 29, 1945, in association with *Allotrichoma alium* Cresson, *Gymnopa* sp. and *Hecamede* sp. (Ephydridæ).

Formosina sp.—Calicoan Island: June 29, 1945, numerous on taro and other vegetation growing in sand in the open along the Pacific Ocean; and July 23, in association with *Aphis medicaginis* Koch, on leguminous plant by the sea.

Prohippelates pallidus (Loew.)—Calicoan Island, June, 1945, in association with *Hecamede albicans* (Meigen) (Ephydridæ).

Undet. sp.—Calicoan Island, August 26, 1945, swept from morning-glory (Convolvulaceæ).

COELOPIDÆ

Coelopa sp.—Calicoan Island, September 6, 1945, from tender foliage of *Thespesia populnea.*

DOLICHOPODIDÆ

Sciapus sp.—Calicoan Island, May, 1945.

DROSOPHILIDÆ

Drosophila, 2 spp., one prob. *melanogaster* Meigen—Calicoan Island, August 10, 1945, from fermenting banana plant.

EMPIDÆ

Drapetis, 2 spp.—Calicoan Island, August 25, 1945, swept from morning-glory (Convolvulaceæ).

EPHYDRIDÆ

Allotrichoma alium Cresson—Calicoan Island, July 29, 1945, in association with *Eutropha* n. sp., near *noctilus* (Walker) (Chloropidæ), and *Gymnopa* sp. and *Hecamede* sp. (Ephydridæ).

Gymnopa sp.—Calicoan Island, July 25, 1945, in association with *Desmometopa* sp. (Ayromyzidæ) and *Hecamede* sp., prob. *persimilis* Hendel (Ephydridæ) from dead land crab on sand; and July 25, from bare sand.

Hebecnema sp.—Calicoan Island, July 23, 1945, in association with *Tethina* sp. (Agromyzidæ) on seaweed along shore of the Pacific Ocean.

Hecamede albicans (Meigen)—Calicoan Island, June, 1945, in association with *Prohippelates pallidus* (Loew.) (Chloropidæ).

H. sp.—Calicoan Island, July 25, 1945, in association with *Desemometopa* sp. (Agromyzidæ) and *Gymnopa* sp. (Ephydridæ) on dead land crab; and July 30 from bare sand.

FUNGIVORIDÆ

Lycoria sp.—Calicoan Island, July 4, 1945, combed from a trapped rat.

LUXANIIDÆ

Homoneura ochripennis (Frey)—Calicoan Island, October 14, 1945, from foliage of lemon seedling in bloom. The flies were easily captured without net.

H. padangensis (de Meijere)—As above.

MUSCIDÆ

Dichaetomyia quadrata (Wd.)—Calicoan Island, August 10, 1945, from fermenting banana plant.

Musca sorbens Wd.—Calicoan Island, May, 1945.

M. vetustissima Walker—Calicoan Island, October 6, 1945, indoors.

Ophyra chalcogaster (Wied.)—Samar, October 7, 1945, from citrus foliage.

Siphona exigus (de Meijere)—Leleboon Island, June 25, 1945, from cow.

Stomoxys calcitrans Linnæus—As above.

Telostylus sp., prob. *decemnotatus* Hendel—Calicoan Island, August 10, 1945, from fermenting banana plant.

OTITIDÆ

Elassogaster metallicus Bigot—Calicoan Island, June, 1945, from vegetation.

Naupoda platessa Osten Sacken—Calicoan Island, October 15, 1945, from bird excrement on jungle foliage.

Scelostenoplerina sp.—Calicoan Island, May, 1945.

PHORIDÆ

Megaselia sp., prob. *scalaris* (Loew.)—Calicoan Island, spring of 1945.

PIOPHILIDÆ

Piophila latipes Meigen—Samar, October 7, 1945, from citrus foliage.

SARCOPHAGIDÆ

Sarcophaga albiceps Meigen—Calicoan Island, June 27, 1945, from jungle, 500 feet above sea level.

S. antilope Bott.—Calicoan Island, May, 1945.

S. knabi Parker—Calicoan Island, August 9, 1945, from *Urena lobata.*

S. misera Walker—Calicoan Island, May and June, 1945.

S. orchidea Bott.—Calicoan Island, May and August, 1945.

S. orientalis Park.—Calicoan Island, June, 1945.

S. orientaloides S. W.—Calicoan Island, May, 1945.

S. sp.—Samar, October 7, 1945, from citrus foliage.

STRATIOMYIDÆ

Merosargus sp.—Calicoan Island, August 10, 1945, from fermenting banana plant.

Negritomyia consobrina (Bigot)—Calicoan Island, October 15, 1945.

Ptilocera smaragdina Walker—Calicoan Island, June, 1945.

Rosapha habilis Walker—Calicoan Island, October 8, 1945, from foliage of *Barringtonia asiatica* along Leyte Gulf.

SYRPHIDÆ

Baccha sp.—Calicoan Island, May to August, 1945.

Tubifera sp.—Calicoan Island: June, 1945; and October 8, 1945, from *Hibiscus tiliaceus.*

Volucella sp.—Samar, May 6, 1945, associated with the psyllid *Mesohomotoma hibisci* (Froggatt) on *Hibiscus tiliaceus;* and Calicoan Island, September 26, from *H. tiliaceus.*

TABANIDÆ

Tabanus sp., near *effilatus* S. S.—Calicoan Island, July 23, 1945, indoors.

TENDIPEDIDÆ

Tendipes sp.—Calicoan Island: June, 1945, numerous on leaves of banana; August 29, at light.

TEPHRITIDÆ

Acidoxantha sp.—Calicoan Island, September 25, 1945, reared from a maggot found feeding in flower bud of *Hibiscus tiliaceus* (September 8). Two other adults of the same species reared from maggots found in buds of the same plant on the same day (September 8) emerged September 27 and 30. From another maggot of the same material, the hymenopterous parasite *Opius longicaudatus* (Ashmead) emerged instead of the fly. Maggots of *Acidoxantha* sp. were found in the flower buds of *Hibiscus tiliaceus* from Samar, also, September 10, but no adults reared.

TYLIDÆ

Grallopoda galbula (Osten Sacken)—Guiuan, Samar, April, 1945, from *Hibiscus tiliaceus* infested with the psyllid *Mesohomotoma hibisci* (Froggatt) and from *Abelmoschus moschatus;* and Calicoan Island, June, 1945, from other vegetation.

G. morbida (Osten Sacken)—Guiuan, Samar, April, 1945, from *Abelmoschus moschatus;* and Calicoan Island, June, 1945, from other vegetation.

HEMIPTERA

ANTHOCORIDÆ

Cardiastethus sp., near *rugicollis* Champ.—Calicoan Island, June, 1945, from pasteboard box containing dry buds of *Hibiscus tiliaceus.*

BELOSTOMATIDÆ

Sphaerodema rusticum (Fabricius)—Calicoan Island, May 21, 1945, dead specimen, from swamp.

COREIDÆ

Cletus sp.—Calicoan Island, May 24, 1945.

Homoeocerus bipustulatus Stål—Calicoan Island, May, 1945.

Leptocorisa acuta (Thunberg)—Calicoan Island, May, 1945.

Physomerus oedimerus (Burmeister)—Calicoan Island, May to September, 1945, from foliage of *Hibiscus tiliaceus* and from other vegetation. Eggs were laid in clusters of 50 to 75 on upper sides of foliage of shrubs and trees of various species. An adult was usually perched on the eggs. A leaf bearing a cluster of 70 eggs and an adult female perched on the eggs was taken indoors for observation. The adult (without being caged) remained constantly on the eggs for six days (August 27 to September 1) and would have probably remained there until the eggs hatched, if she had not been severely disturbed by transfer of the material. The eggs hatched nine days after having been abandoned by the adult (September 10), indicating an incubation period of 15 days or longer.

Riptortus linearis (Fabricius)—Calicoan Island, May, 1945.

R. pedestris Stål—Calicoan Island, June, 1945.

GERRIDÆ

Limnogonus sp.—Samar, April, 1945.

HYDROMETRIDÆ

Hydrometra lineata Eschscholtz—Calicoan Island, June, 1945, from brackish water.

LYGAEIDÆ

Astacops nigripes Stål—Calicoan Island, October 18, 1945, numerous on tree trunk, 400 feet above sea level.

A. sp.—Leleboon Island, June 25, 1945, from foliage.

Dasynus coccocinctus Burmeister—Leleboon Island, June 25, 1945, rare.

Dieuches uniguttatus (Thunberg)—Calicoan Island, August 10, 1945, from jungle vegetation.

Geocoris flaviceps (Burmeister)—Calicoan Island, May and July, 1945.

MIRIDÆ

Hyalopeplus vitripennis Stål—Calicoan Island, June, 1945, from foliage.

Pachypeltis ståli Distant—As above.

PENTATOMIDÆ

Antestia cruciata (Fabricius)—Calicoan Island, October 10, 1945, from foliage of a shrub along seashore of Leyte Gulf.

Chrysocoris germari var. *consul* (Vollenhoven)—Calicoan Island, May 13, 1945, from jungle vegetation.

Cuspicona sp.—Calicoan Island, June 7, 1945.

Cyclopelta obscura (Lepelletier & Serville)—Calicoan Island, August, 1945.

Eysarcoris bovillus Dallas—Calicoan Island, May and June, 1945.

E. guttigerus Thunberg—As above.

E. sp.—Calicoan Island, May and August, 1945, from jungle vegetation.

Undet. sp. of tribe Acanthosomini, probably a new genus near *Cyphostethus* Fieber—Calicoan Island, October 15, 1945, from shrub bearing berries, 350 feet above sea level, only one other specimen was seen. It was from a plant of the same species.

PLATASPIDÆ

Coptosoma cincta (Eschscholtz)—Leleboon Island, May, 1945, from legume (sonting).

PYRRHOCORIDÆ

Dysdercus crucifer Stål—Calicoan Island, May to October, 1945, feeding on flower buds, seed pods, and foliage of *Hibiscus tiliaceus*, apparently its preferred host.

D. megalopygus Breddin—Calicoan Island, Leleboon Island, and Samar, April to October, 1945, from *Urena lobata*, *Sida* spp., and *Abelmoschus moschatus*.

D. poecilus (Herrich-Schäffer)—Same localities, dates, and hosts as, and usually in association with, *D. megalopygus*.

REDUVIIDÆ

Endochus histrionicus Stål—Calicoan Island, May, 1945.

Euagoras tagalicus Stål—Leleboon Island, June 23, 1945, eggs, nymphs, and adults, on shrub along seashore.

E. sp.—Calicoan Island, May, 1945.

Rihirbus trochantericus Stål—Calicoan Island, May, 1945.

Stachyomerus pallescens Stål—Calicoan Island, August 10, 1945, from jungle.

Sphodronyttus erythropterus (Burmeister)—Calicoan Island, May, 1945.

S. semirufus Stål—Calicoan Island, June 27, 1945, 500 feet above sea level.

Sycanus ståli Dohrn.—Calicoan Island, May and June, 1945.

Veledella sp.—Calicoan Island, May, 1945.

Vesbius purpureus Thunberg—Calicoan Island, July 30, 1945, indoors.

Undet. sp., apparently of a new genus close to *Epidaus*—Calicoan Island, May, 1945.

HOMOPTERA

APHIDÆ

Aphis citricidus (Kirkaldy)—Samar and Calicoan Island, April and May, 1945, from citrus foliage.

A. fabæ Scopoli—Calicoan Island, May, 1945, probably from an herbaceous legume (sonting).

A. laburni Kaltenbach—Calicoan Island, June and July, 1945, from two species of legume, sonting and another.

CERCOPIDÆ

Phymatostetha montana Schmidt—Calicoan Island, June, 1945.

CICADELLIDÆ

Bothrogenia sp. near *ferruginea* (Fabricius)—Calicoan Island, May 7, 1945.

Cicadella sp.—Calicoan Island, May, 1945.

Tartessus malayus Stål—Calicoan Island, May, 1945.

CICADIDÆ

Cosmopsaltria inermis Stål—Samar, spring of 1945.

COCCIDÆ

Lepidosaphes belonging to the *tubulorum*-complex—Calicoan Island, June 27, 1945, on leaves of a jungle tree 400 feet above sea level.

Pinnaspis sp.—Leleboon Island, June 25, 1945, on foliage of shrub along seashore.

Pseudococcus lilacinus (Cockerell) ?—Calicoan Island, October 14, 1945, from tree in brackish swamp adjacent to Leyte Gulf.

P. (*Ferrisia*) *virgatus* (Cockerell)—Leleboon Island, June 25, 1945, on guava foliage and twigs; and Samar, spring, 1945, on citrus and *Codiaeum variegatum.*

Saissetia hemisphaerica (Targiori-Tozetti)—Calicoan Island, May 15, 1945, on underside of leaves of a jungle shrub.

DELPHACIDÆ

Delphacodes sp.—Calicoan Island, August 25, 1945, at light.

Liburnia furcifera Horváth—As above.

FLATIDÆ

Mesophylla alba Jac.—Calicoan Island, May 24, 1945.

FULGORIDÆ

Dictyophara, 2 spp., one prob. *nakanonis* Matsumura—Calicoan Island and Samar, May to September, 1945.

Epora subtilis Walker—Calicoan Island, May, 1945.

Mindura sp.—Calicoan Island, October 14, 1945, from vegetation in dense jungle.

Neomelicharia calichroma (Walker)—Leleboon Island, June 29, 1945, numerous on breadfruit.

Virgilia sp., prob. new—Calicoan Island, May, 1945.

MEMBRACIDÆ

Gargara nigrocarinata Funkhouser—Samar, August 29, 1945, from foliage of *Hibiscus tiliaceus.*

G. nitidipennis Funkhouser—As above.

G. varicolor Stål—Calicoan Island, May to October, 1945.

Tricentrus pilinervosus Funkhouser—Samar, April, 1945, from *Abelmoschus moschatus.*

PSYLLIDÆ

Mesohomotoma hibisci (Froggatt)—Guiuan, Samar, April, 1945, from *Hibiscus tiliaceus.*

Tenaphalara fascipennis (Crawford)—Calicoan Island, October 15, 1945, from leaves of a rubberlike plant.

HYMENOPTERA

ANTHOPHORIDÆ

Anthophora sp.—Calicoan Island, September 11, 1945, from foliage in swamp.

APIDÆ

Apis dorsata Fabricius, the so-called giant or wild honeybee, "wild" referring to the fact it cannot be domesticated—Calicoan Island, May 8, 1945, at light.

A. florea Fabricius—Calicoan Island, August 10, 1945, found dead on jungle foliage.

Thyreus sp.—Calicoan Island, June, 1945.

BRACONIDÆ

Campyloneurus sp.—Calicoan Island, May, 1945.

Iphaulax sp.—As above.

Microbracon sp., apparently new—Calicoan Island, June, 1945.

Opius longicaudatus (Ashmead)—Calicoan Island, September 27, 1945, emerged from puparium of *Acidoxantha* sp.; period of development 20 days or longer (notes under *Acidoxantha* sp., Diptera, Tephritidæ).

Spathius sp.—Calicoan Island, May, 1945.

ENCYRTIDÆ

Psyllæphagus sp.—Guiuan, Samar, April, 1945, from *Hibiscus tiliaceus* infested with *Mesohomotoma hibisci* (Froggatt) (Psyllidæ).

FORMICIDÆ

Anoplolepis longipes (Jerdon)—Calicoan Island: June, 1945, attending *Aphis laburni* Kaltenbach on legume; June 25, 1945, a nuisance in kitchen; and September 7, on tips of twigs of *Thespesia populnea.*

Camponotus (*Colobopsis*) sp.—Calicoan Island: May 8, 1945, at light; May 13, from foliage of *Hibiscus tiliaceus;* August 29 and September 5, at light; September 7 from tips of twigs of *Thespesia populnea;* October 14, from shrub along seashore of Leyte Gulf. And Samar, September 3, 1945, attending a species of mealybug (*Pseudococcus*) on fruit of *Ficus* sp.

Crematogaster sp.—Calicoan Island: May 10, 1945, from foliage of *Hibiscus tiliaceus;* and May 15, attending *Saissetia hemisphærica* (Targioni-Tozetti) on underside of leaves of a jungle shrub.

Diacamma sp.—Calicoan Island, June, 1945, one carrying a mutilated homopteron.

Dolichoderus (*Hypoclinea*) *bituberculatus* (Mayr.)—Samar, August 29, 1945 and Calicoan Island, September 9, from foliage of *Hibiscus tiliaceus.*

Monomorium (*Lampromyrmex*) sp.—Calicoan Island, May 8, 1945.

Odontoponera transversa (F. Smith)—Calicoan Island, September 9, 1945, from sand in the open.

Oecophylla smaragdina (Fabricius)—Calicoan Island, August 5, 1945, from jungle vegetation.

Paratrechina longicornis (Latreille)—Calicoan Island: September 9, 1945, from sand in the open; and October 15, 1945, from flower buds of a shrub along seashore of Leyte Gulf.

Polyrhachis cyaniventris (F. Smith)—Calicoan Island, May and June, 1945.

P. ypsilon Emery—As above.

Solenopsis geminata rufa (Jerdon)—Calicoan Island: August 15 and 23, 1945, as household pest at different places on the island; and September 9, from foliage of *Hibiscus tiliaceus* and from sand in the open.

ICHNEUMONIDÆ

Theronia sp.—Calicoan Island, August 19, 1945, from fermenting banana plant in jungle.

MEGACHILIDÆ

Megachile sp.—Calicoan Island, August 12, 1945.

MELIPONIDÆ

Trigona sp.—Calicoan Island, May, July, and August, 1945.

PSAMMOCHARIDÆ

Batazonus orientalis (Cameron)—Guiuan, Samar, September 8, 1945, from foliage of *Urena lobata.*

SCOLIIDÆ

Campsomeris aureicollis (Lepeletier)—Calicoan Island, August 9, 1945, outdoors; and August 27, indoors.

C. sp.—Calicoan Island, May, 1945.

SPHECIDÆ

Argogorytes sp.—Calicoan Island, August 24, 1945, indoors.

Chlorion aurulentus sericeus (Fabricius)—Calicoan Island, October 9, 1945, indoors.

C. hæmorrhoidalis muticus (Kohl)—Calicoan Island, August 29, 1945.

C. hæmorrhoidalis siamensis (Taschenberg)—Calicoan Island, May, 1945.

C. luteipennis (Mocsary)—Calicoan Island, September 9, 1945, from sand in the open.

C. umbrosa plumifera (Costa)—Calicoan Island, September 11, 1945, from foliage in swamp.

Lyroda venusta Bingham—Calicoan Island, August 24, 1945, swept from morning-glory (Convolvulaceæ).

STEPHANIDÆ

Stephanus sp.—Calicoan Island, May, 1945.

VESPIDÆ

Polistes dubius de Saussure—Calicoan Island, May, 1945.

Rygchium atrum de Saussure—Calicoan Island and Samar, September, 1945.

XYLOCOPIDÆ

Xylocopa sp.—Calicoan Island, May and June, 1945.

ISOPTERA

TERMITIDÆ

Nasutitermes (*N.*) *panayensis* Oshima—Calicoan Island, June, 1945, indoors.

ODONATA

LIBELLULIDÆ

Erythrodiplax sp.—Calicoan Island, May, 1945, from jungle swamp.

Sympetrum sp.—As above.

ORTHOPTERA

BLATTIDÆ

Blattella germanica (Linnæus)—Calicoan Island, April to October, 1945, household pest.

Epilampra sp.—Calicoan Island, June, 1945, indoors.

Panesthia sp.—As above.

Symploce sp.—Calicoan Island, October 18, 1945, from jungle, 400 feet above sea level.

Undet. sp. of Pseudomopinæ—Calicoan Island, June, 1945, from *Hibiscus tiliaceus* in swamp and September 10, from other vegetation.

PHASMATIDÆ

Sipyloidea, 2 spp.—Calicoan Island and Leleboon Island, June, 1945, from jungle vegetation.

LOCUSTIDÆ

Catantops infuscatus (De Haan)—Calicoan Island, May, 1945.
Oxya sp.—Calicoan Island, August, 1945.

MANTIDÆ

Hierodula patellifera (Serville)—Calicoan Island, May, 1945.
Leptomantis sp.—As above.

TETTIGONIIDÆ

Anerota sp.—Calicoan Island, July 26 and August 25, 1945.

LEPIDOPTERA

AMATIDÆ

Amata (?) sp.—Calicoan Island, summer of 1945.
Callitomis sp.—As above.

COSMOPTERYGIDÆ

Pyroderces, prob. n. sp.—Calicoan Island, June, 1945, reared from dry seed pods of *Hibiscus tiliaceus.*

GELECHIIDÆ

Pectinophora gossypiella (Saunders)—Calicoan Island, September 17, 1945, larvæ from flower buds of *Thespesia populnea.*

GLYPHIPTERYGIDÆ

Tortyra sp.—Calicoan Island, June 26, 1945.

NYMPHALIDÆ

Hypolimnas antilope (Cramer)—Calicoan Island, June, 1945, reared from caterpillars on *Morus* sp. in jungle.

PHALAENIDÆ

Undet. sp.—Calicoan Island, September 9, 1945, immature larva feeding in young seed pod of *Hibiscus tiliaceus.*

PHYCITIDÆ

Undet. sp.—Calicoan Island, October 8, 1945, caterpillars within web defoliating *Barringtonia asiatica* along Leyte Gulf.

PYRALIDÆ

Diaphanea sp.—Calicoan Island, May, 1945, at light.

PYRAUSTIDÆ

Dichocrocis surusalis (Walker)—Calicoan Island, June 21, 1945, emerged from caged flower buds and seed pods of *Hibiscus tiliaceus;* September 8 to 12, many larvæ of this species or some other of the family were taken feeding in flower buds, blooms and young seed pods of the same host (*H. tiliaceus*), but no adults reared.

XYLORCTIDÆ

Undet. sp.—Calicoan Island, Samar of 1945, larvæ feeding in flower buds and seed pods of *Hibiscus tiliaceus.*

SIPHONAPTERA

PULICIDÆ

Ctenocephalides felis (Bouche)—Calicoan Island, July 23, 1945, from dog.

Pulex irritans Linnæus—Calicoan Island: June 25, 1945, from man; and October 10, from dog.

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ARTIFICIAL FERTILIZATION AND EMBRYOLOGY OF MIROGOBIUS LACUSTRIS HERRE

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TWO PLATES

This paper presents notes on the artificial fertilization and the early development of *Mirogobius lacustris* Herre, a small transparent goby of the family Gobiidæ. Roxas and Blanco (1937) made a revision of the genus *Mirogobius* Herre (Gobiidæ) based on the constant vertebral and the greater fin ray counts of the two known species *M. lacustris* and *M. stellatus*. *M. lacustris* is known as *dolong* in Tagalog, and *kip-kip* in Iloko. It is found in Lanigay, Polangui, Albay Province; Laguna de Bay, Laguna Province; and Paoay Creek, Paoay and Butong Lake, Laoag, Ilocos Norte Province. It is a source of goby fry used for food.

Artificial fertilization.—The artificial fertilization of the kip-kip was undertaken in August, 1939, as a contribution to the early life histories of Philippine fresh-water fishes. Sexually mature females of *M. lacustris* are easily recognized by the presence of ripe, intermediate, and immature eggs in their transparent bodies. Males of the species are larger than the females; their heads are larger and bulldoglike, and the genital organs, decidedly larger.

The following procedure was followed in artificial fertilization: A ripe female was removed from an aquarium with a small dipnet; its abdomen was pressed gently towards its genital opening with the thumb and forefinger. As a result of the pressure eggs sprung from the oviduct one at a time. The eggs extruded were placed in a clean watch glass with a fine pincer. Each egg is provided with long adhesive threads that radiate from the apical poles. The eggs were attached to one another by means of these adhesive threads, to form clusters. Adhesive threads or filaments of eggs are morphological characteristics of cyprinids, atherinids, and phallostethids. The filaments or threads protect the eggs during embryonic development by

keeping them intact and protecting them from being drifted by currents and other physical agencies. Hence, egg filaments are necessary for pelagic eggs that require a longer time for development.

A dissection of a ripe female was made to ascertain the type of eggs in the ovary. The immature eggs (Plate 1, fig. 1) are oblong and nucleated. The intermediate eggs are more or less globular with a quantity of yolk material (Plate 1, fig. 2). A mature egg, which is about 1 mm in diameter (Plate 1, fig. 3), carries a much greater amount of yolk material and its perivitelline space is narrower in the yolk-sphere.

A sexually mature male was also removed from the aquarium, and its abdomen also gently pressed towards its genital opening. The pressing was done in such a way that the milt dropped on the eggs which were placed in the watch glass half filled with water from the aquarium. The artificially fertilized eggs were later transferred to two watch glasses containing tap water which was changed daily. The incubation period of the eggs under laboratory conditions lasted from three to four days.

Embryology of M. lacustris.—An observation of the embryological development of the fertilized egg was made with the aid of a compound miscroscope, and all drawings of the living materials were made with the aid of a camera lucida.

About thirty minutes after fertilization the egg shell changes its globular shape into a pear-shaped appearance (Plate 1, fig. 4). First cleavage is very apparent in the yolk sphere by the presence of a blastodisc protoplasm of about equal the size of the yolk of the egg.

One hour after fertilization the blastodisc divides into equal daughter cells (Plate 1, fig. 5). About one and a half hours after fertilization the second plane of cleavage appears cutting the first plane at right angles (Plate 1, fig. 6). The blastodisc of eight cells has a bilateral symmetry two and a half hours after fertilization (Plate 1, fig. 7). The multiplication of the cells after this stage is very irregular until the mass of protoplasm of the blastodisc covers one-half of the yolk sphere (Plate 1, figs. 8–9). Twenty hours after fertilization the germ ring is developed (Plate 1, figs. 10–11). A group of cells are pushed in towards the cleavage cavity thus forming the embryonic shield (Plate 1, fig. 12). As the blastoderm increases rapidly in size and the germ ring advances around the yolk, the embryonic shield has grown larger and more de-

finitely outlined as a linear thickening on the anteroposterior axis of the former embryonic shield (Plate 2, fig. 1). The later embryonic stages are very much noticeable when the embryo increases in size and the yolk sphere diminishes in size. An embryo coiled around more than half of the yolk sphere (Plate 2, fig. 2) has the beginning of the eyes thirty hours after fertilization. The optic vesicles and eight somites are developed thirty-six hours after fertilization (Plate 2, figs. 3–4). Embryonic circulation is in evidence forty-eight hours after fertilization (Plate 2, fig. 5). The embryo has developed fin folds and the yolk is very much reduced in size sixty-four hours after fertilization (Plate 2, fig. 6). The embryo is very active within the egg shell and changes its position every other minute. Plate 2, fig. 7, is an illustration of embryo in the shell seventy-eight hours after fertilization. Viewed dorsally eighty hours after fertilization, the embryo shows well-developed head, eyes, ear bones, reduced yolk sac, and traces of larval intestines and myotomes (Plate 2, fig. 8). The newly hatched larva (Plate 2, fig. 9), eighty-four hours after fertilization, has a well-developed notochord which does not extend to the axial lobe of the caudal fin; the dorsal fin fold is as narrow as that of the ventral fin; the myotomes are well developed. Traces of the larval intestine which runs parallel the notochord and behind the reduced yolk sac are apparent. The head has well-developed eyes and ear bones.

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ILLUSTRATIONS

[Camera lucida drawings by the author.]

PLATE 1. MIROGOBIUS LACUSTRIS HERRE

FIG. 1. Immature egg; × 100.
2. Intermediate egg; × 100.
3. Mature egg, top view; × 100.
4. Egg, one-cell stage; × 100.
5. Egg, two-cell stage; × 100.
6. Egg, four-cell stage; × 100.
7. Egg, eight-cell stage; × 100.

FIGS. 8–9. Eggs showing multiplication of cells; × 100.
10–11. Eggs showing germ ring and blastula stages; × 100.

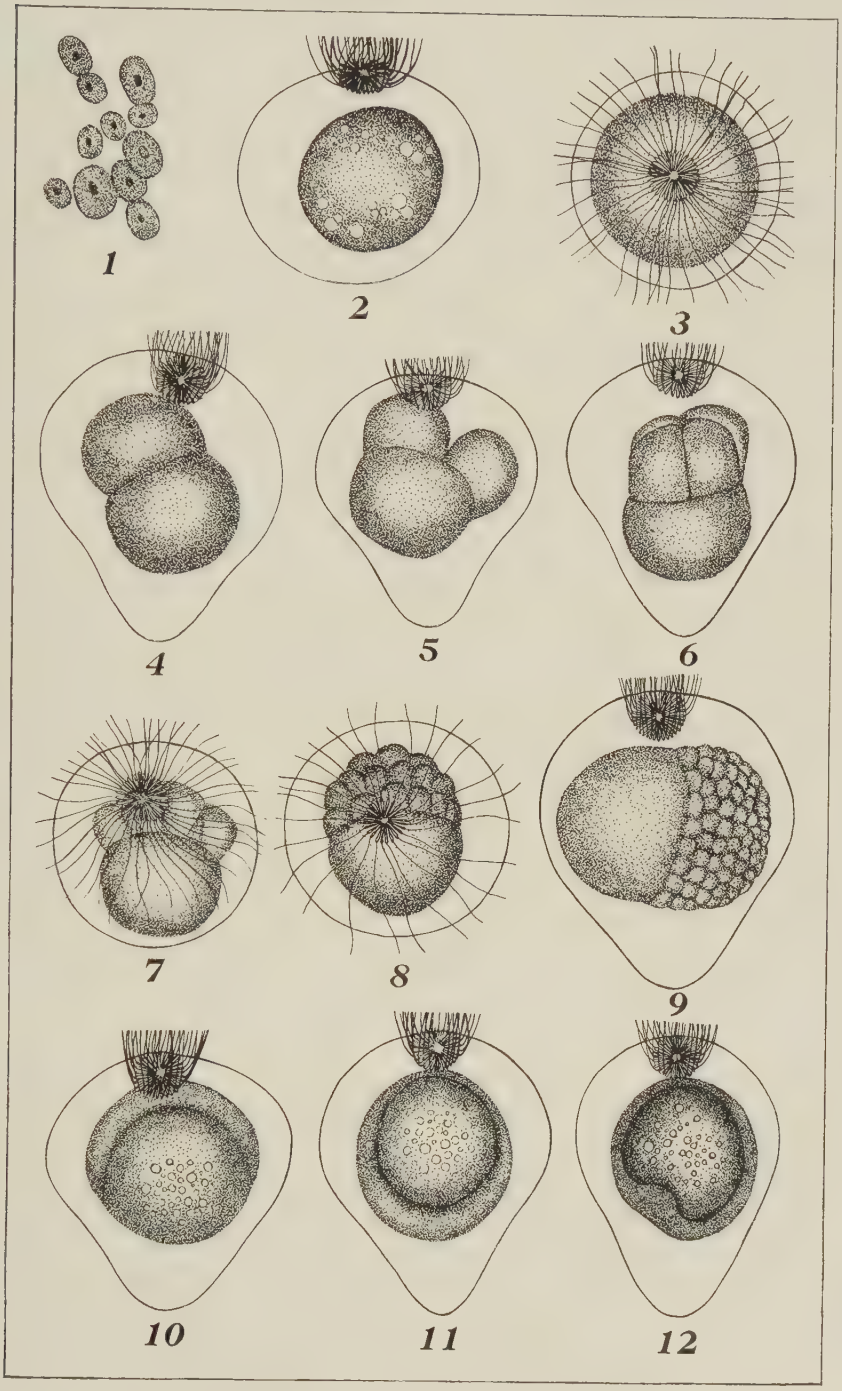
FIG. 12. Egg showing embryonic shield; × 100.

PLATE 2. MIROGOBIUS LACUSTRIS HERRE

FIG. 1. Egg showing primitive streak; × 100.
2. Egg showing developing embryo; × 100.

FIGS. 3–4. Embryo, 36 hours after fertilization; × 100.

FIG. 5. Embryo, 48 hours after fertilization; × 100.
6. Embryo, 64 hours after fertilization; × 100.
7. Embryo, 78 hours after fertilization; × 100.
8. Embryo, 80 hours after fertilization; × 100.
9. Larva, 84 hours after fertilization; × 100.



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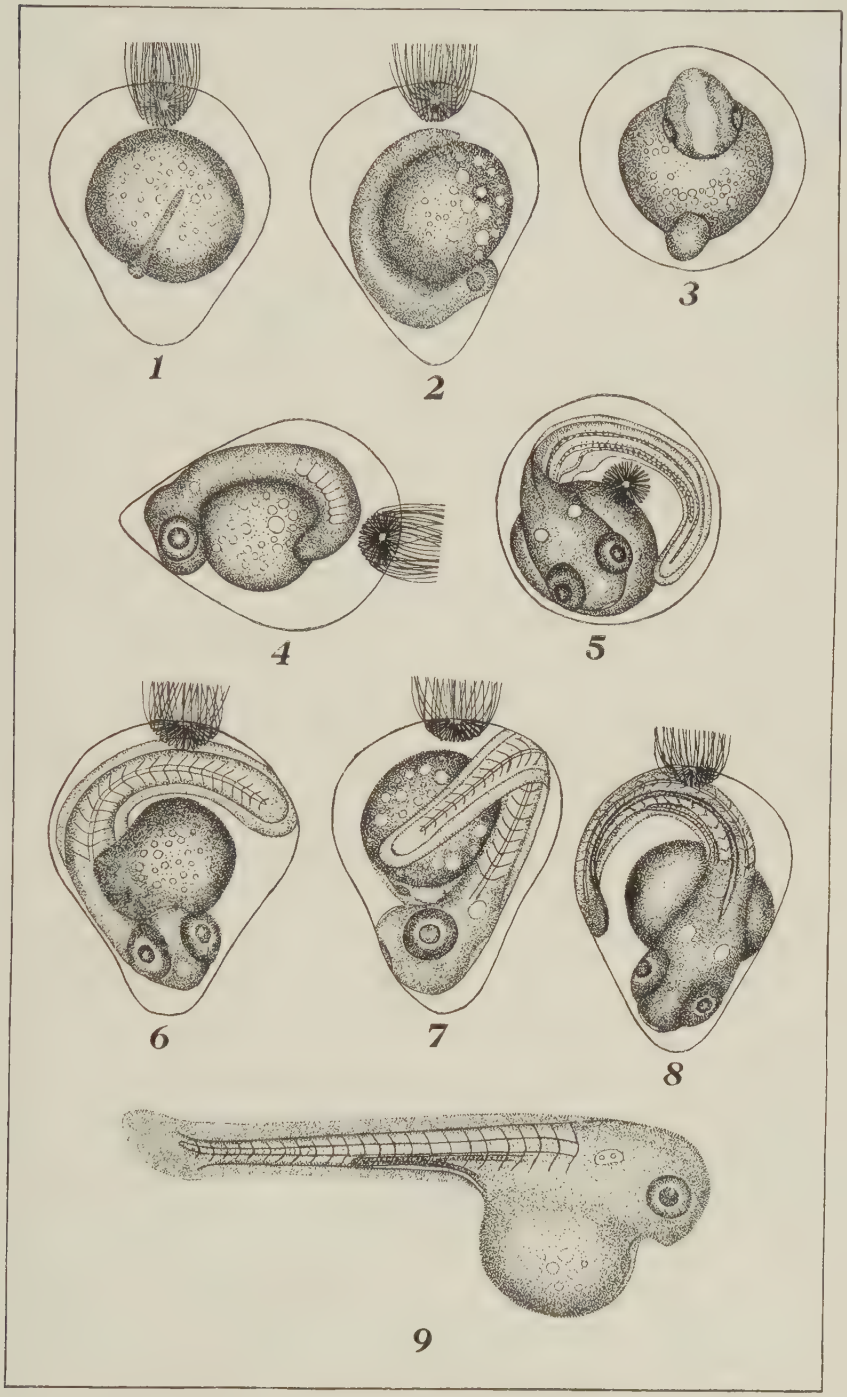

PLATE 2. MIROGOBIUS LACUSTRIS HERRE.

THE BREEDING ACTIVITIES AND EMBRYOLOGY OF APLOCHEILUS LUZONENSIS HERRE AND ABLAN

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THREE PLATES

Aplocheilus luzonensis Herre and Ablan, a cyprinodont, is known among the Ilocanos as *coscosleng*. It abounds in rivers, streams, ponds, and ditches of the municipalities of Solsona, Batac, Laoag, Bacarra, and Dingras, Ilocos Norte Province. This fresh-water fish is generally not caught for food, but during scarcity of food fish it is taken advantage of by the inhabitants, especially those of the town of Solsona. This fish is voracious, feeding largely on mosquito larvæ, plankton, and organic detritus that float along littoral margins of shallow ponds and streams. Its flat head and transverse mouth are characteristic adaptations to surface feeding habits. Aside from its importance as a mosquito exterminator it may be kept as lively aquarium fish. Its small size and beautiful golden-yellow color at the proximal edges of the dorsal, anal, and caudal fins, especially during the breeding season, make it an attractive ornamental fish of distinct value.

Breeding activities.—Since the discovery of coscosleng as a new species of the family Cyprinodontidæ by Herre and Ablan in 1934, field study on the extent of its distribution and on the occurrence of its larvæ and young stages has been carried on. *Aplocheilus luzonensis* is known to breed throughout the year, but the height of the breeding season occurs in August. The coscosleng is in the habit of swimming in slow-running waters along littoral margins of ponds or streams where there is abundant vegetation of vallisneria, anacharis or other aquatic plants. This species in great numbers invariably congregate in water one to three feet deep. The males and females are not nest builders. On the other hand the eggs of females are provided with egg filaments. So far as known, such egg filaments are also developed in the developing egg of Atherinidæ, Phallostethidæ, and Gobiidæ.

The female of the species is recognized by the bulging of the flunk around the pectoral fins. The female is usually smaller

than the male, the latter having a larger head and a brighter golden-yellow color on the caudal and dorsal fins.

Breeding females usually carry clusters of eggs hanging in their oviduct. The outer egg membranes have numerous short adhesive threads and also a group of long filamentous threads arising from an area of the egg membrane. Such long filamentous threads are twisted and join other twisted threads of other eggs to form a single cord (Plate 1, fig. 1). A female which is ready to spawn is unusually active because she is being pursued by breeding males. When the female is ready to extrude eggs she becomes less active, preferring to settle at the gravelly bottom of a margin of a stream, rubbing off her abdomen on the gravel or peebles. She lies on a dorsolateral position at times followed by caudal fin vibrations until the eggs are extruded one at a time. A gravid female carries from 5 to 28 eggs (Table 1) depending upon the size of the female fish. Fertilization of the eggs is external as it was observed that ripe males followed females with extruded clusters of eggs. Clusters of eggs which are fertilized are either carried by the females until they are hatched or detached from the oviduct of the female fish and then attached to some plant leaves until they are hatched. In nature fertilized clusters of eggs which are not detached from the oviduct of the parent fish have more chances of being aërated, protected, and hatched than those clusters of eggs detached from the oviduct. Such eggs may be devoured by carnivorous fishes or other aquatic predatory species.

TABLE 1.—*Number of ripe ova in A. luzonensis.*

Length of fish in mm.	Number of eggs per fish	Length of fish in mm.	Number of eggs per fish
32	28	25	14
31	28	24	9
29	24	22	5
30	26	23	7
28	17	21	5
27	26	20	7
26	20	19	5

The breeding activities of this fresh-water cyprinodont appear to be characteristically different from those of other fresh-water species owing to the lack of copulatory external organs, as those found in the members of the family Phallostethidæ. The courtship prior to the spawning activity is not very apparent as that of the fresh-water species, which are nest-builders.

Aside from the field observations on the breeding activities of the coscosleng, the behavior of gravid females and adult males was also observed in a glass aquarium to facilitate the embryological study of *A. luzonensis.*

Embryology of A. luzonensis.—Clusters of eggs detached from the oviduct of the female fish were removed from aquatic plants and then transferred to watch glasses. Water from the aquarium was used daily up to the time of hatching. The observations and drawings were made with the aid of a camera lucida on all living materials. The incubation period of *A. luzonensis* in August, 1939, lasted from eight to ten days depending upon laboratory conditions.

The newly laid but unfertilized egg is transparent, about 1.5 mm in diameter, not globular, and has a narrow perivitelline space (Plate 1, fig. 2). The perivitelline space becomes wider a few hours after fertilization. One hour after fertilization the blastodisc (Plate 1, fig. 3) is apparently well differentiated, appearing as a protrusion of protoplasm at the pole of the yolk sphere. The oil globules are reduced in number and also occupy the mid portion of the yolk sphere. One and a half hours after fertilization meridional cleavage takes place (Plate 1, fig. 4), the blastodisc dividing into two equal daughter cells. About two and a half hours after fertilization the second plane of cleavage is apparent (Plate 1, fig. 5), thus cutting the first cleavage at right angles and dividing the blastodisc into four equal cells. After the eight cell-stage, cell division of the blastoderm was observed to be variable (Plate 1, fig. 6). The blastoderm continues to increase in diameter (Plate 2, fig. 1) until it covers a third of the yolk sphere. Twenty-five hours after fertilization the original primitive streak is very much advanced (Plate 2, fig. 2). Plate 2, fig. 3, shows a developing embryo forty-eight hours after fertilization. The embryo has developed eyes. Fifty-two hours after fertilization (Plate 2, fig. 4) the developing embryo has thirteen somites. An embryo, seventy-two hours old (Plate 2, figs. 5–6), has eighteen somites. At this stage the embryonic circulation is very much advanced; the notochord is very distinct; and the ear bones and brain are already in evidence, on the way to development.

The yolk sphere undergoes reduction, the number of somites increases to twenty-five, and the embryonic circulation is more advanced than in an embryo seventy-six hours after fertilization (Plate 3, fig. 1). One hundred hours after fertilization

the embryo as shown dorsally (Plate 3, fig. 2) has well-developed large eyes and ear bones. The pulsating heart, the smaller yolk-sphere, and the more or less continuous finfold are very much noticeable in the embryo one hundred twenty-four hours old (Plate 3, fig. 3). Seven days after fertilization (168 hours) the embryo begins to hatch by breaking the eggs shell through the process of wriggling inside the egg wall and finally hatching, tail first (Plate 3, fig. 4). The larva at the age of two days measures 5 mm long and has a well-developed pectoral and a single median fin that starts dorsally about the middle of the back and around the notochord up to the ventral surface. The larva has dark stellate pigment spots on the sides of the body (Plate 3, fig. 5).

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ILLUSTRATIONS

[Camera lucida drawings by the author.]

PLATE 1. APLOCHEILUS LUZONENSIS HERRE AND ABLAN

FIG. 1. Cluster of eggs; × 300.
2. A ripe egg; × 600.
3. An egg one hour after oviposition showing blastodisc; × 600.

FIGS. 4–6. Eggs showing multiplication of cells 3 to 4 hours after fertilization; × 600.

PLATE 2. APLOCHEILUS LUZONENSIS HERRE AND ABLAN

FIG. 1. Egg, 8 hours after fertilization; × 600.
2. Egg, 25 hours after fertilization showing advance primitive streak; × 600.
3. Developing embryo, 48 hours after fertilization; × 600.
4. Developing embryo, with thirteen somites, 52 hours after fertilization; × 600.

FIGS. 5–6. Embryos, 72 hours after fertilization, stages of embryo with 13–18 somites; × 600.

PLATE 3. APLOCHEILUS LUZONENSIS HERRE AND ABLAN

FIG. 1. Embryo, 76 hours after fertilization; × 600.
2. Embryo, 100 hours after fertilization; × 600.
3. Embryo, 124 hours after fertilization; × 550.
4. Embryo, 168 hours after fertilization; × 550.
5. Larva, 192 hours after fertilization; enlarged.

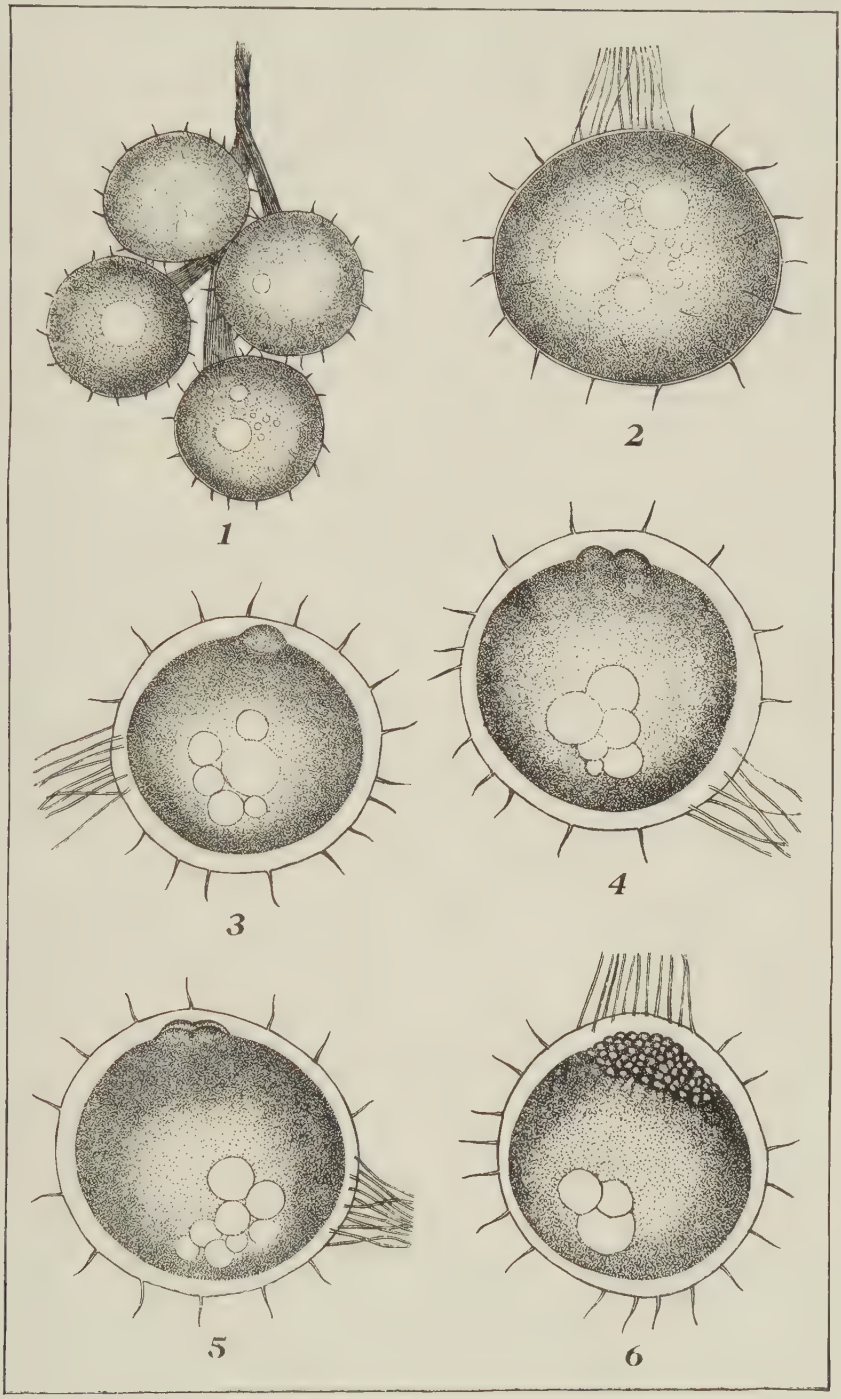
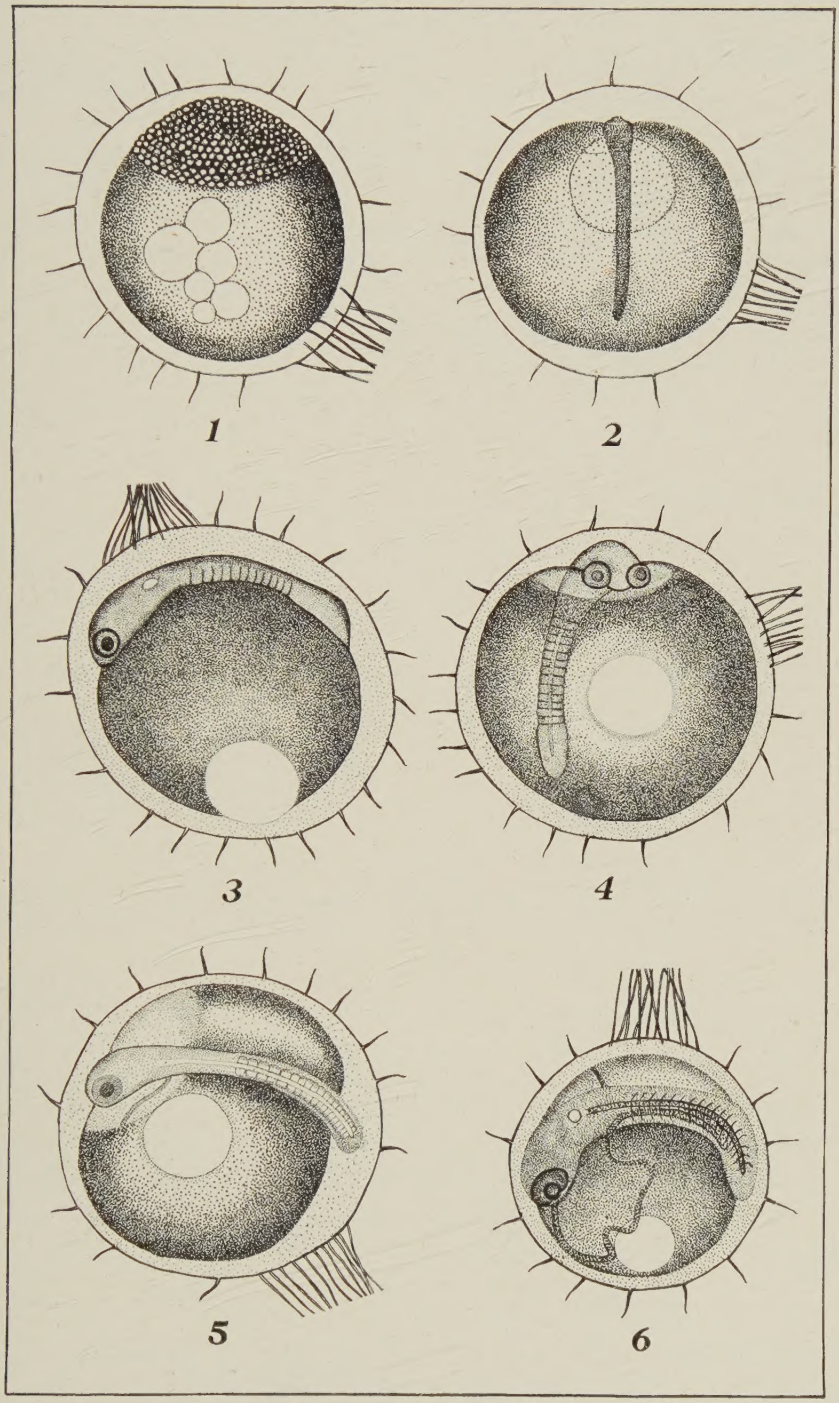


PLATE 1. APLOCHEILUS LUZONENSIS HERRE AND ABLAN.



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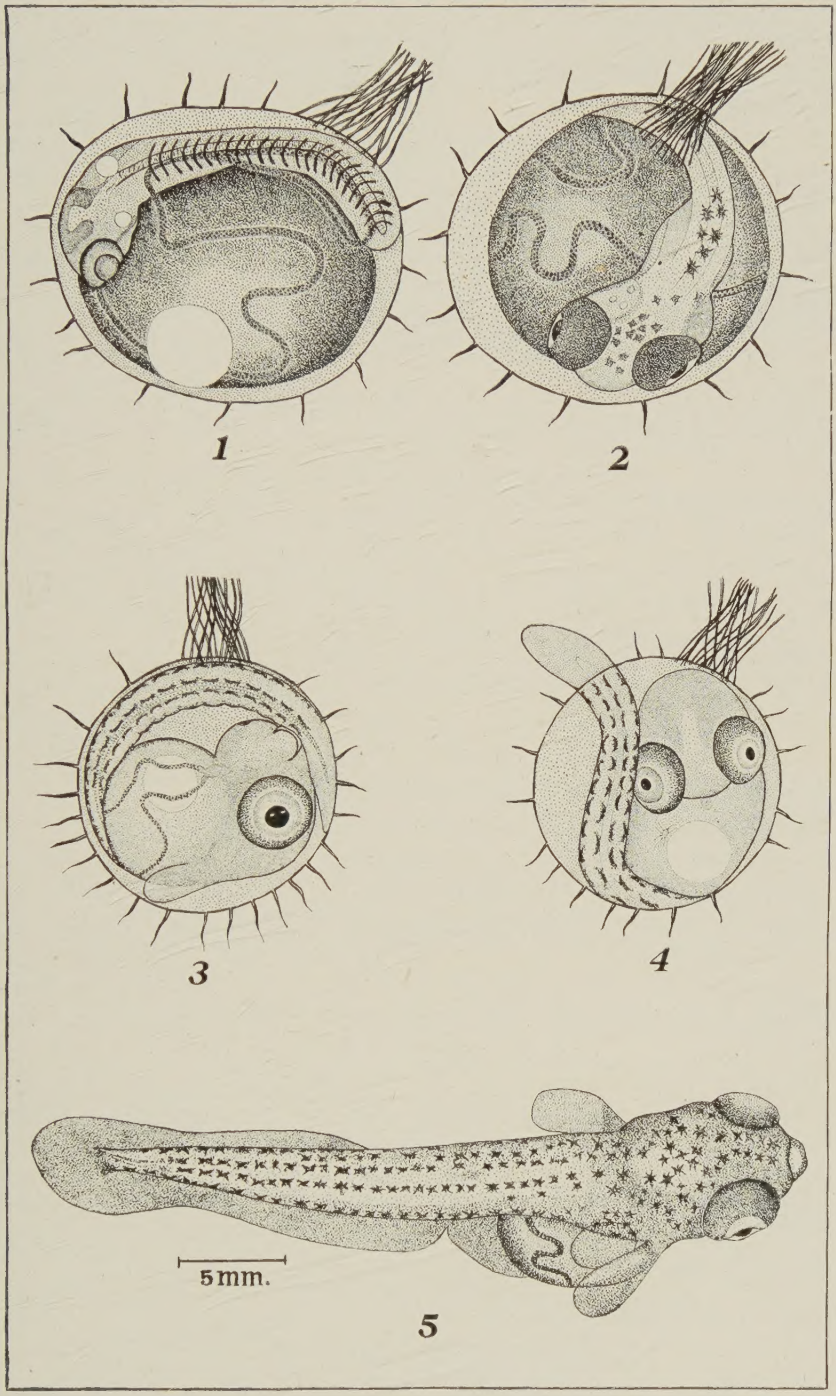

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